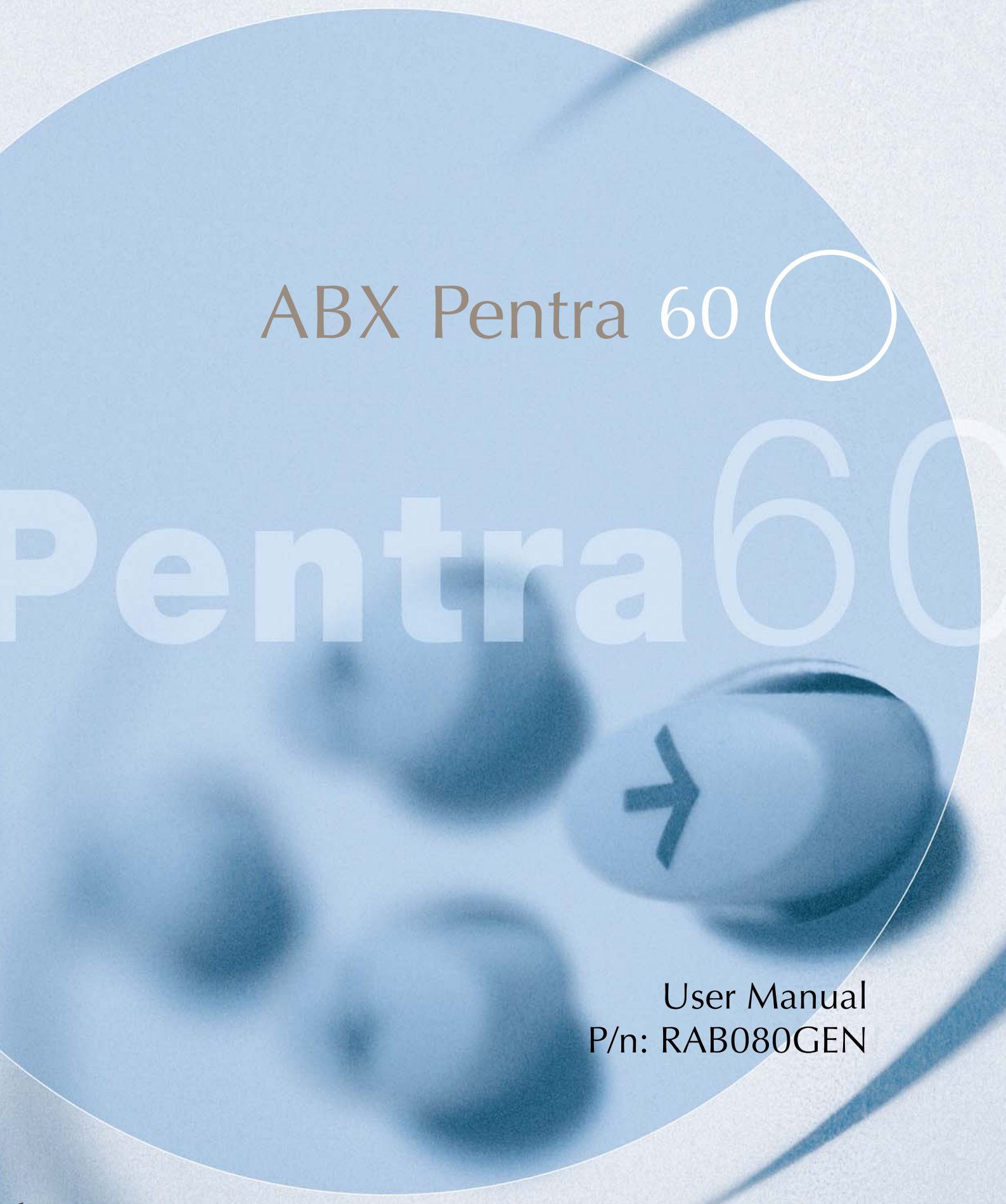
ABX Pentra 60



User Manual
P/n: RAB080GEN

HAN 618A

HORIBA ABX
Diagnostics

Instrument User Manual Update

RAM207AEN

ABX Pentra 60
User Manual Update

Please, take note of the modifications on next pages. Please, cross out the appropriate sections in the user manual prior to inserting this addendum at the beginning of the user manual.

Date: 25/09/06

FORM 0860 - rev 1

User Manual Update

Tab.1-1: Concerned sections of the ABX Pentra 60 user manual RAB080G

Section	Page	Paragraph	Item change
Specifications	1-17	5.3. Known interfering substances	Known interferences due to chemotherapy Interferences in the basophil count
Specimen Run & Results	3-6	4. Running specimens	Recommendations on the analysis mode
	3-26	5.3.3. Flags on WBC/BASO histogram	L1 Flag
	3-29	5.3.6. WBC balance	CBC mode limitations (WBC Balance)

1. WBC Balance

The WBC balance flags (LMNE+ and LMNE-) are activated only if the test selected is «DIFF» and if this flag has been activated. The WBC Balance can be enabled or disabled by an approved HORIBA ABX Service Technician. Contact your local HORIBA ABX Technical Service Representative for selection of this option.

These flags are associated with an (!) on all differential parameters (% and #).

The WBC balance flag will indicate an instrument defect or it can also highlight a known interference (see User manual).
In the case of pathology whose treatments weaken the leucocytic membranes, the agent of lysis of WBC channel can damaged the cells and give a lower leukocytes counting.
The LMNE+ flag will then be triggered off and a suspicion will be integrated to the WBC results. We thus recommend not to disable WBC balance flag and to work in DIFF mode for all the samples which can present this possible interference. Selecting the CBC mode will disable this control mode. It is thus recommended to use this mode for patient not presenting this type of interference.

2. L1/LL1 Flag

In certain cases, the L1 flag will not be triggered off because of the poor sensitivity of this flag (large platelet aggregates and/or erythroblasts that are beyond the electronic threshold). This happens in CBC mode only. Two additional flags LL (see User manual) and LL1 (see User manual) are available in DIFF mode and provide more reliability in anomaly detection. This mode should be recommended.

3. Recommendations on the analysis mode selection (CBC or DIFF)

◆ When selecting CBC analysis, there is no control mode on WBC erroneous countings that may be caused by specific treatments on patients (see User manual) and WBC balance description (see User manual).

4. Known interfering substances

4.1. WBC

Chemotherapy: Cytotoxic and immunosuppressive drugs may increase the fragility of the leukocyte membranes which may cause low WBC counts. In these particular cases, CBC mode must not be used as WBC balance alarm (see User manual) is disabled. It is recommended to run these samples in DIFF mode.

4.2. Basophil

Over evaluation in the Basophil count

◆ Excessive number of leukocytes (leukocytosis) can cause artificial rise in the number of counted basophils due to the shifting of the leukocyte population in the zone of the basophil one.
◆ Monocytes and Blasts show large granules and may shift on the basophil counting area. This may interfere with an accurate count.
◆ An abnormally low number of leukocytes (leukopenia) may increase too the basophil results. The elements present in the zone of basophil are brought back on a small total quantity of leukocytes, which increases the statistical error and may cause variabilities in the percentage.
◆ The weakness of leukocyte cells shown in certain diseases (Chronic Lymphocytic Leukemia) or during anti-cancer treatment (chemotherapy) can be translated on the basophilic channel by under evaluation of the leukocytes because of their destruction and thus cause a statistical increase in the basophil ones.

Under evaluation in the Basophil count

◆ During leukemia, basophils may lose their cytochemical characters and react abnormally with the reagent. The destruction of the basophil cytoplasms prevents their differentiation with the other leukocytes.
◆ The basophils with very small sizes (following treatments) may interfere with leukocyte counts, as cell sizes can not be distinguished.
◆ The abnormal basophils (degranulation following allergies) may interfere with leukocyte counts, because cell sizes can not be distinguished and because they may lose their characteristic intracytoplasmic material.

ABX Pentra 60

User Manual

P/n: RAB080GEN

CE IVD

HORIBA ABX
B.P. 7290
34184 MONTPELLIER Cedex 4 - FRANCE

HORIBA ABX
Diagnostics

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REVISIONS

INDEX	TECHNICAL NOTE	REVISION	SECTIONS	DATE
A		CREATION	ALL	15/06/1999
B	RAH670AA	BASOPHIL FLAG+MISCELLANEOUS CORRECTIONS	ALL	03/11/1999
C	RAH739AA	LINEARITY LIMITS	1+5	24/05/2000
D	RAH764AA	SOFTWARE RELEASE V2.0	ALL	25/09/2000
E	RAH986AA	SOFTWARE RELEASE V2.1.6	ALL	24/09/2003
F	RAN193A	SOFTWARE RELEASE V2.2.0	RAM145A	26/07/2005
G	RAN153B	COMPANY NAME CHANGE	ALL	02/09/2005

- This document applies to the latest software version as indicated above.

- When a subsequent software version changes the information in this document, a new section and/or sections will be released.

- Latest version documents on www.horiba-abx.com.

- Declaration of conformity

Latest version of the CE declaration of conformity for this instrument is available on www.horiba-abx.com.

1. WARNINGS AND PRECAUTIONS

NOTE

NOTE: Emphasizes the important information especially helpful to the operator before, during or after a specific operational function.

IMPORTANT

IMPORTANT: Emphasizes an operating procedure that must be followed to avoid erroneous results.

CAUTION

CAUTION: Emphasizes an information that must be followed to avoid possible damage to the instrument or erroneous results.

WARNING

WARNING: Flags a procedure that if not followed properly, can prove to be extremely hazardous to either the operator or the environment or both.

1.1. Warnings

- User manual must be entirely read and personnel trained by *HORIBA ABX* before attempting to operate instrument. The user always operates with full knowledge and appreciation of instrument warnings, alarms and flags.
Always refer to labeling and *HORIBA ABX* instructions in order to avoid to compromise system integrity.

- The *ABX PENTRA 60* responds to the Standards and Directives named in the declaration of conformity.

- The reagents and accessories stipulated by *HORIBA ABX* have been validated in accordance with the European Directive for in-vitro medical devices (98/79/CE).

- The use of any other reagents and accessories may place at risk the performance of the instrument, engaging the Users reponsability. In this case, *HORIBA ABX* takes no responsability for the device nor for the results rendered.

- Disposal gloves, eyes protection and lab coat must be worn by the operator. Local or national regulations must be applied in all the operations.

- Portable/mobile should not be used in proximity of the instrument.

- All peripheral devices should be IEC compatible.

1.2. Limited garantee

• The duration of guarantee is stipulated in the Sales conditions associated with the purchase of this instrument. To validate the guarantee, ensure the following is adhered to:

1 - The system is operated under the instructions of this manual.

2 - Only software or hardware specified by *HORIBA ABX* is installed on the instrument. This software must be the original copyrighted version.

3 - Services and repairs are provided by an *HORIBA ABX* authorized technician, using only *HORIBA ABX* approved spare parts.

4 - The electrical supply of the laboratory follows the national regulations.

5 - Specimens are collected and stored in normal conditions.

6 - Reagents used are those specified in this user manual.

7 - Proper tools are used when maintenance or troubleshooting operations are performed.

CAUTION

If this instrument has been supplied to you by anyone other than HORIBA ABX or an authorised representative, HORIBA ABX cannot guarantee this product in terms of specification, latest revision and latest documentation. Further information may be obtained from your authorised representative.

1.3. Safety precautions, electronic and moving parts

• The following parts must not be handled or checked by the user:
- Electrical power supply,
- Electronic components.

• Operator injury may occur from an electric shock. Electronic components can shock and injure the user. Do not tamper with the instrument and do not remove any components (covers, doors, panels and so on) unless otherwise instructed within this document.

• Danger! The battery may explode if it is not replaced correctly! Replace only with the same or equivalent type recommanded by the manufacturer. Dispose of used batteries according to the manufacturer's instructions.

• Moving parts:
It is strictly forbidden to disable sensors as it may cause operator injuries. Protection covers must not be opened during instrument operations.

1.4. Biological risks

• Consider all Specimens, Reagents, Calibrators, Controls, etc… that contain human blood or serum as potentially infectious! Use established, good laboratory working practices when handling specimens. Wear protective gear, Gloves, Lab coats, Safety glasses and/or Face shields, and follow other bio-safety practices as specified in OSHA Blood borne Pathogens Rule (29 CFR part 1910. 1030) or equivalent bio-safety procedures.

• *HORIBA ABX* uses disinfectant product for instrument decontamination and highly recommends to decontaminate regularly your instrument.

1.5. Instrument cleaning

Instrument external cleaning

• The external surfaces of the instrument must be decontaminated considering the biological environment.

WARNING

Never spill liquid on the instrument.

Never use Disinfectant product* that contains alcohol.

• Screen: Use a soft clot, slightly wet with disinfectant product*. Wipe gently the screen and dry to remove any trace of moisture.

• All contaminated surfaces (covers, counting assembly area...): Slightly wet a sponge with disinfectant product* and wipe the dirty surfaces.

• Stainless steel parts: Slightly wet a sponge with disinfectant product* and wipe the dirty surfaces. Dry with a soft cloth.

* Products having the following microbiological properties:
• Bactericidal
• Fungicidal
• Active on Aspergillus fumigatus
• Active on Mycobacterium tuberculosis (B.K)
• Antiviral (VIH, HBV and rotavirus)

Product Example validated by *HORIBA ABX*:
ANIOS detergent disinfectant ; *WIP'ANIOS ; ref: 1316.424*

NOTE

Please also refer to the W.H.O (World Health Organization) guidelines: «Laboratory Biosafety Manual, 2nd edition», for further information.

Instrument internal cleaning

• Concentrated cleaning: Counting chambers and hydraulics parts are decontaminated by using the «Concentrated cleaning» function as described further in this manual.

Sampling probe

• Sampling probe must be decontaminated as follows:

1- Prepare a solution of Sodium Hypochlorite to 100ml/l.

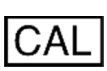
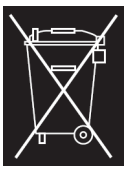
2- Fill a 5ml tube with this solution.

3- Run 5 analysis on bleach.

NOTE

Please also refer to the W.H.O (World Health Organization) guidelines: «Laboratory Biosafety Manual, 2nd edition», for further information.

1.6. Graphics and symbols

	Switch off position		Switch on position
	Alternating current		Manufacturer
	In Vitro Diagnostic medical device		This product conforms to the EEC Standards and Directives named in the declaration of conformity
	Caution, consult accompanying documents		Biological risk
	Reagent		Up
	Fragile, handle with care		Keep dry
	Do not stack		Temperature limitation
	Batch code		Catalogue number
	Use by		Consult Instructions for Use
	Calibrator		Control
	Content		This product should be disposed of and recycled at the end of the useful life in accordance with the WEEE Directive (2002/96/CE)

2. WORKING CONDITIONS

2.1. Environment

• The operation of the *ABX PENTRA 60* should be restricted to indoor location use only! Operation of the instrument at altitudes of over 3000 Meters (9800 feet) is not recommended. The instrument is designed for safety from voltages surges according to INSTALLATION CATEGORY II and POLLUTION DEGREE 2 (IEC EN 61010-1).
Please contact your local *HORIBA ABX* representative for any information regarding the operation location when it does not comply with the recommended specifications.

2.2. Location

• The *ABX PENTRA 60* should be placed on a clean and levelled table or workbench. Please note that the *ABX PENTRA 60*, printer and reagents weigh approximately 40 kilograms (88 lbs).
Avoid exposure to sunlight.
Place your instrument where it is not exposed to water or vapor.
Place your instrument where it is free from vibration or shock.
Place your instrument where an independent power receptacle can be used.
Use a receptacle different from the one used by a device that easily generate noise such as a centrifuge, etc...

• Provide a space of at least 20 cm (8 inches) at the back of the instrument for arranging the power cable and tubings.

• The Power switch and Input voltage supply connection should always be accessible! When positioning the system for operational use, leave the required amount of space for easy accessibility to these items.

2.3. Grounding

• Proper grounding is required when installing the system. Check the wall outlet ground (Earth) for proper grounding to the facilities electrical ground. If you are unsure of the outlet grounding, contact your facilities engineer to verify the proper outlet ground!

2.4. Humidity and temperature conditions

• *ABX PENTRA 60* must operate between temperatures of 16 to 34°C (61 to 93°F). Maximum relative humidity should be 80% for temperatures up to 31°C (88°F) and decreasing linearly to 50% relative humidity at 40°C (104°F). If the system is kept at a temperature of 10°C (50°F) or less, it must be allowed to sit at room temperature for 1 hour before it can be used for operation.

2.5. Electromagnetic environment check

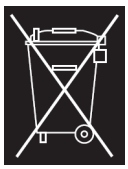
• The *ABX PENTRA 60* has been designed to produce less than the accepted level of electromagnetic interferences in order to operate in conformity with its destination, allowing the correct operation of other instruments also in conformity with their destination.
In case of suspected electromagnetic noise, check that the instrument has not been placed in the proximity of electromagnetic fields or short wave emissions, i. e. Radars, X-rays, Scanners, Cell phones, etc...

2.6. Environmental protection

• Disposal Used accessories and consumables must be collected by a laboratory specialized in elimination and recycling of this kind of material according to the local legislation.

• Disposal *ABX PENTRA 60* instrument:
It should be disposed of in accordance with local legislation and should be treated as being contaminated with blood. The appropriate biological precautions should be taken.

If any doubt, please contact your *HORIBA ABX* representative service department.



• European Legislation:
In accordance with the European Directive (2002/96/CE, known also as WEEE), instruments having this symbol, and sold into a European country by *HORIBA ABX* or an authorised representative must be disposed of and recycled correctly at the end of its useful life. Due to the local changing regulations in each country, please contact your local representative for detailed and upto date information on how to appropriately dispose of the instrument.

2.7. Transport and storage conditions

• Storage temperature: -20°C +50°C

Prior to the shipping of an instrument by transporter, whatever the destination, an external decontamination of the instrument must be carried out.

2.8. Installation

• An *HORIBA ABX* representative will install your instrument, computer, software and printer.

2.9. Package content

• Verify that all of the parts from the package list are present:

ABX PENTRA 60	
1x RAB080XXX	User manual
1x XAA473A	EPSON LX300+ printer
1x	Pentra 60
1x DAC0XXA	Supply cable
1x XEA791A	Installation kit

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1. TECHNICAL SPECIFICATIONS

• The *ABX PENTRA 60* is a fully automated hematology analyzer used for in vitro diagnostic testing of whole blood specimens.

• The *ABX PENTRA 60* is able to operate either in **CBC** mode (**C**ell **B**lood **C**ount: 12 parameters) or in **CBC + 5DIFF** mode (**5** population **Diff**erential count: 26 parameters).

1.1. Parameters

1.1.1. CBC Mode

WBC	White Blood Cell
RBC	Red Blood Cell
Hgb	Hemoglobin concentration
Hct	Hematocrit
MCV	Mean Corpuscular Volume
MCH	Mean Corpuscular Hemoglobin
MCHC	Mean Corpuscular Hemoglobin Concentration
RDW	Red Distribution Width
Plt	Platelets
PDW *	Platelet Distribution Width
MPV	Mean Platelet Volume
Pct *	Plateletcrit

**** PDW and PCT have not been established as indications for this product, in the United States. The use of PCT and PDW should be restricted to research and Investigational measurements only.***

1.1.2. CBC + 5DIFF Mode

WBC	White Blood Cell
LYM	Lymphocytes % and #
MON	Monocytes % and #
NEU	Neutrophils % and #
EOS	Eosinophils % and #
BAS	Basophils % and #
LIC *	Large Immature Cell % and #
ALY *	Atypical Lymphocyte % and #
RBC	Red Blood Cell
Hgb	Hemoglobin concentration
Hct	Hematocrit
MCV	Mean Corpuscular Volume
MCH	Mean Corpuscular Hemoglobin
MCHC	Mean Corpuscular Hemoglobin Concentration
RDW	Red Distribution Width
Plt	Platelets
PDW *	Platelet Distribution Width
MPV	Mean Platelet Volume
Pct *	Plateletcrit

PCT, PDW, ALY and LIC have not been established as indications for this product, in the United States. The use of PCT, PDW, ALY and LIC should be restricted to research and Investigational measurements only.

1.1.3. Units

UNITS							
	WBC	RBC	HGB	HCT	PLT	MCV	
STD	$10^3/mm^3$	$10^6/mm^3$	g/dl	%	$10^3/mm^3$	μm^3	
SI	$10^9/l$	$10^{12}/l$	g/l	l/l	$10^9/l$	fl	
mmol/l	$10^9/l$	$10^{12}/l$	mmol/l	l/l	$10^9/l$	fl	
	MCH	MCHC	RDW	MPV	PCT	PDW	
STD	pg	g/dl	%	μm^3	%	%	
SI	pg	g/l	%	fl	$10^{-2}/l$	%	
mmol/l	fmol	mmol/l	%	fl	$10^{-2}/l$	%	
	LYC	LYC	MON	MON	NEU	NEU	EOS
STD	%	#	%	#	%	#	%
SI	%	#	%	#	%	#	%
mmol/l	%	#	%	#	%	#	%
	EOS	BAS	BAS	ALY	ALY	LIC	LIC
STD	#	%	#	%	#	%	#
SI	#	%	#	%	#	%	#
mmol/l	#	%	#	%	#	%	#

- To select the set of units, refer to *Chapter 4. Instrument configuration*.

1.2. Throughput Analyses

- CBC Mode (CBC): 60/h
- CBC + 5DIFF Mode (DIF): 60/h

1.3. Tube Identification

- By means of the worklist or identification file.
- By means of the Barcode labels.

1.4. Reagents

- *ABX* ***DILUENT*** (20 litres),
- *ABX* ***CLEANER*** (1 litre, Integrated),
- *ABX* ***EOSINOFIX*** (1litre, Integrated),
- *ABX* ***BASOLYSE II*** (1 litre, Integrated),
- *ABX* ***ALPHALYSE*** or ***LYSEBIO*** (0.4 litre, Integrated).

1.5. Keyboard & Display

- *HORIBA ABX* specific keyboard, silicone made.
- LCD, 128 x 240 pixels, LED Backlighted.

1.6. Data processing

- 68331 type microprocessor.
- RS232C output.

1.7. Measurements and computation

- Impedance for WBC, Plt, RBC, BASO.
- Photometry for Hgb.
- Impedance and light scattering for LYM, MON, NEU, EOS, ALY and LIC.
- Computation from stored data that was directly measured for Hct, MCV, MCH, MCHC, RDW, MPV, Pct, PDW.

2. PHYSICAL SPECIFICATIONS

2.1. Power requirements

- Power supplies: ..... from 100Vac to 240Vac (±10%)
  ..... 50 Hz to 60 Hz
- Power Consumption: .... Analyzer: 200 VA
  ..... Printer (Depends on the printer, see Printer's manual.)

2.2. Operating temperature and humidity

- 16 - 34°C (61 - 93°F) room temperature.
- Maximum relative humidity 80% for temperatures up to 31°C (88°F) decreasing linearly to 50% relative humidity at 40°C (104°F).

2.3. Dimensions and Weight

- Analyzer dimensions: ... Height: approximately 516 mm (20.3 in.)
  ..... Width: approximately 444 mm (17.5 in.)
  ..... Depth: approximately 481 mm (19 in.)
- Analyzer weight: ..... approximately 35Kgs (77lbs)

2.4. Minimum specimen volume

- CBC Mode (CBC): ..... * 30µl
- CBC + 5DIFF Mode (DIF): ..... * 53µl

2.5. Dilution ratios

- WBC/BASO 1/200
- LMNE 1/80
- RBC/Plt 1/10 000
- Hgb 1/250

2.6. Hgb measurement

- Hgb chamber, LED 555 nm.
- Modified Drabkin method (cyanmethemoglobin)

- Light source ..... Electroluminescent diode
- Wavelength ..... 550nm $\pm$ 10nm

2.7. Counting aperture diameters

- WBC/BASO ..... 80 µm
- LMNE ..... 60 µm
- RBC/Plt ..... 50 µm

2.8. Reagent consumption (ml)

Cycle	Reagent					Approximate Duration
	Diluent	Basolyse II	Cleaner	Eosinofix	Alphalyse	
CBC Cycle	20.4	2.1	0.9	x	0.4	60”
Diff Cycle	25.6	2.1	0.9	1	0.4	60”
Startup*	60.8	2.1	3.7	1	1.4	3’53”
Shutdown	20.5	x	14	x	1	2’48”
Prime Diluent	42.9	x	x	x	x	3’03”
Prime Cleaner	1.1	x	24.8	x	x	1’22”
Prime Eosinofix	1.6	x	x	23.6	x	1’12”
Prime Basolyse II	1.1	23.6	1.1	x	x	1’20”
Prime alphalyse	2.1	x	x	x	8.4	1’27”
Prime all reagents	47	24	25.1	24	8.2	6’
Autoclean cycle	14.2	1	1	1	0.3	1’38”
Autocontrol cycle	23.4	x	1.4	x	1	1’4”
RBC chamber cleaning	2.5	x	x	x	x	7”
Unprime all	x	x	x	x	x	6’25”
Chambers rinsing	12.6	1	1	1	0.3	1’17”
Cytometer rinsing	4.9	x	x	x	x	1’11”
Concentrated cleaning	25	x	1.4	x	0.9	4’10”
Cleaning	12.6	1	1	1	0.3	1’19”

**** for one background count only (maxi = 3)***

3. SUMMARY OF PERFORMANCE DATA

3.1. Precision (Reproducibility)*

- The instrument was initially calibrated with Minocal Calibrator (Lot No. CX322).

- Three levels of ABX MINOTROL material (Lot No: JX108) were run in duplicate once daily for a prolonged period on all parameters.
The results were used to quantify within run precision, and Total Precision in accordance with the NCCLS EP 5-A Guidelines.

Parameter	ABX Minotrol Control	Within run SD	SD of run means	SD of daily means	Total Precision (SD)
WBC	JX108 Low	0.001	N/A	N/A	0.001
	JX108 Normal	0.008	N/A	N/A	0.013
	JX108 High	0.031	N/A	N/A	0.045
RBC	JX108 Low	0.000	N/A	N/A	0.000
	JX108 Normal	0.001	N/A	N/A	0.001
	JX108 High	0.002	N/A	N/A	0.003
HGB	JX108 Low	0.001	N/A	N/A	0.001
	JX108 Normal	0.002	N/A	N/A	0.007
	JX108 High	0.005	N/A	N/A	0.012
HCT	JX108 Low	0.021	N/A	N/A	0.025
	JX108 Normal	0.064	N/A	N/A	0.122
	JX108 High	0.104	N/A	N/A	0.181
PLT	JX108 Low	6.271	N/A	N/A	20.646
	JX108 Normal	40.229	N/A	N/A	72.103
	JX108 High	154.146	N/A	N/A	381.388
Parameter	**ABX Minotrol Control**	**Within run CV%**	**CV% of run means**	**CV% of daily means**	**Total Precision (CV%)**
WBC	JX108 Low	1.8	N/A	N/A	1.93
	JX108 Normal	0.9	N/A	N/A	1.12
	JX108 High	0.9	N/A	N/A	1.05
RBC	JX108 Low	0.8	N/A	N/A	0.86
	JX108 Normal	0.6	N/A	N/A	0.79
	JX108 High	0.7	N/A	N/A	0.88
HGB	JX108 Low	0.4	N/A	N/A	0.57
	JX108 Normal	0.3	N/A	N/A	0.59
	JX108 High	0.4	N/A	N/A	0.60
HCT	JX108 Low	0.9	N/A	N/A	0.99
	JX108 Normal	0.7	N/A	N/A	0.97
	JX108 High	0.7	N/A	N/A	0.89
PLT	JX108 Low	3.1	N/A	N/A	5.69
	JX108 Normal	2.6	N/A	N/A	3.46
	JX108 High	2.5	N/A	N/A	4.01

* Source: 510K submission K030144.

Precision claims*

Parameter	% CV	Nominal Values
WBC	< 2.0%	10.0 x 10^3/mm^3
RBC	< 2.0%	4.67 x 10^6/mm^3
HGB	< 1.0%	13.6 g/dl
HCT	< 2.0%	36.0 %
PLT	< 5.0%	243 x 10^3/mm^3

* Source: 510K submission K030144.

3.2. Precision (Repeatability)

• Based on 10 consecutive samples of the same fresh whole blood sample without alarm:

Parameters	%CV	Test level
WBC	<2%	at 10x10^3/mm^3
RBC	<2%	at 5x10^6/mm^3
Plt	<5%	at 300x10^3/mm^3
Hgb	<1%	at 15 g/dL
Hct	<2%	at 45%

3.3. Linearity limits*

• **Linearity range**: The Manufacturer's tested linearity zone of the instrument using linearity kits and/or human blood.

• **Linearity limits**: Maximum and minimum values within instrument returns no dilution alarm.

• **Visible range**: Range values given by the instrument. These values (above linearity limits) are given as an indication. They are given associated with a «D» flag. This Visible range is outside Manufacturer's range.

• **Linearity kits**: Linearity was tested using available «Low Range» and «Full Range» Linearity Test kits. The Test kits were analyzed and data was computed according to the Manufacturer's instructions.

• **Human Blood**: Linearity was also obtained on human blood, using a minimum of 5 dilution points. The results of this study are as follows:

Parameter	Linearity Range	Linearity Limits	Visible Range	Error Limit
WBC (10^3/µl)	0.40 to 130.80	0 to 120	120 to 150	±0.3 ±7.5%
RBC (10^6/µl)	0.23 to 9.76	0 to 8	8 to 18	±0.07 ±3%
HGB (g/dl)	0 to 31.06	0 to 24	24 to 30	±0.3 ±3%
HCT (%)	1.80 to 88.90	0 to 67	67 to 80	±2 ±3%
PLT (10^3/µl) (for Hgb≥2g/dl)	3.30 to 2007	0 to 1900	1900 to 2800	±10 ±12.5%
PLT (10^3/µl) (for Hgb<2g/dl & Plt>15x10^3/mm^3)	7 to 2895	0 to 2800	2800 to 3200	±10 ±12.5%

* Source: 510K submission K030144.

3.4. Carry-over*

• The *ABX PENTRA 60* carry-over effects were evaluated by assaying a sample with high cell concentrations three consecutive times (i1-3), followed immediately by testing a diluted sample consecutively 3 times (j1-3).

$$\text{Carry-over} = \frac{(j1-j3)}{(i3-j3)} X\ 100$$

Carry-over % is then:

	WBC	RBC	HGB	PLT
Mean low levels	1.06	1.58	5.28	31.33
Mean high levels	58.81	6.37	22.03	1106.67
Carry-over %	-0.260	0.000	-0.179	-0.186

This is the method as described in *Guidelines for the Evaluation of blood cell analyzers including those used for differential leukocyte and reticulocyte counting and cell marker applications. ISLH, 14 January, 1994.*

• Carry-over Conclusion:
Results provided are extremely satisfactory. In order to provide for eventual possibilities within the laboratory environment the following claims shall be made:

	WBC	RBC	HGB	PLT
Claims	< 2.0%	< 2.0%	< 2.0%	< 2.0%

* Source: 510K submission K030144.

3.5. Normal ranges*

Parameters	Male	Female
WBC (10^3/µL)	4 - 10	4 - 10
RBC (10^6/µL)	4.50 - 6.50	3.80 - 5.80
HGB (g/dL)	13.0 - 17.0	11.5 - 16.0
HCT (%)	40.0 - 54.0	37.0 - 47.0
MCV (µm^3)	80 - 100	80 - 100
MCH (pg)	27.0 - 32.0	27.0 - 32.0
MCHC (g/dL)	32.0 - 36.0	32.0 - 36.0
RDW (%)	11.0 - 16.0	11.0 - 16.0
PLT (10^3/µL)	150 - 500	150 - 500
MPV (µm^3)	6 - 11	6 - 11
PCT (%)	0.15 - 0.50	0.15 - 0.50
PDW (%)	11 - 18	11 - 18
NEU (%)	50 - 80	50 - 80
LYM (%)	25 - 50	25 - 50
MON (%)	2 - 10	2 - 10
EOS (%)	0 - 5	0 - 5
BAS (%)	0 - 2	0 - 2

IMPORTANT

Expected values will vary with sample population and/or geographical location. It is highly recommended that each Laboratory establish its own Normal ranges based upon the local population!

* Bibliography:
AIDE MEMOIRE D'HEMATOLOGIE
Prof : C.SULTAN / M. GOUAULT- HELMANN / M. IMBERT
Service Central d'Hématologie de l'Hopital Henri Mondor
Faculté de médecine de Créteil (Paris XII)

3.6. Accuracy*

• The data shows good correlation between results achieved on the *ABX PENTRA 60* versus the reference system, which can be resumed as follows:

Parameter	R^2(Comparison of means)	Accuracy Claims
WBC	0.997	>0.95
PLT	0.998	>0.95
RBC	N/A	N/A
HGB	N/A	N/A
HCT	N/A	N/A
LYM	N/A	N/A
NEU	N/A	N/A
MON	N/A	N/A
EOS	N/A	N/A
BAS	N/A	N/A

* Source: 510K submission K030144.

3.7. Leukocyte differential count

• Not available at the time of publication.

3.8. Sample stability study

• Not available at the time of publication.

3.9. Results exceeding instrument capacities

RESULTS DISPLAYED			
PARAMETER	LINEARITY LIMITS	VISIBLE RANGE	> VISIBLE RANGE
WBC	«result»	«result+D»	DIL
RBC	«result»	«result+D»	DIL
HGB	«result»	«result+D»	DIL
HCT	«result»	«result+D»	DIL
PLT (for Hgb≥2g/dl)	«result»	«result+D»	DIL
PLT (for Hgb<2g/dl & Plt>15x10³/mm³)	«result»	«result+D»	DIL

RESULTS TRANSMITTED OR PRINTOUT			
PARAMETER	LINEARITY LIMITS	VISIBLE RANGE	> VISIBLE RANGE
WBC	«result»	«result+D»	--.-- + D
RBC	«result»	«result+D»	--.-- + D
HGB	«result»	«result+D»	--.-- + D
HCT	«result»	«result+D»	--.-- + D
PLT (for Hgb≥2g/dl)	«result»	«result+D»	--.-- + D
PLT (for Hgb<2g/dl & Plt>15x10³/mm³)	«result»	«result+D»	--.-- + D

NOTE

Use the instrument diluent to dilute the sample if a ---- D flag occurs on WBC or Hct.

- Results displayed and printed out:

«PLT-C» message indicates the triggering of the PLT extended linearity mode for an Hgb<2g/dl & Plt>15x10³/mm³ between 1900x10³/µl and 2800x10³/µl.

- Results transmitted:

«C» message indicates the triggering of the PLT extended linearity mode for an Hgb<2g/dl & Plt>15x10³/mm³ between 1900x10³/µl and 2800x10³/µl.

4. REAGENTS SPECIFICATIONS

4.1. Reagent specifications

• In order for the instrument to operate correctly, high-quality reagents must be used. *HORIBA ABX* provides all the necessary reagents.

NOTE

The HORIBA ABX reagents specified for this instrument have been approved in accordance with the European Directive 98/79/CE (Annex III) for in-vitro medical devices.

• The CD-Rom RAX055 delivered with your instrument provides Reagents, Controls and Calibrators leaflets/msds.
Latest versions of these documents are available on www.horiba-abx.com/documentation.

4.2. Waste handling precautions

WARNING

When disposing of waste, protective clothing must be worn (lab coat, gloves, eye protection, etc…). Follow your local and/or national guidelines for biohazard waste disposal.

• If required, waste can be neutralized before being discarded. Follow your laboratory's protocol when neutralizing and disposing of waste.

• Dispose of the waste container according to the local or national regulatory requirements.

5. LIMITATIONS

WARNING

Whilst every effort is taken by HORIBA ABX to investigate and indicate all known interference's, it is by no means possible to guarantee that all interference's have been identified. At all times, results should be validated and communicated only once all information relating to the patient has been assessed and taken into account.

5.1. Maintenance

• In *Chapter 5. Maintenance & Troubleshooting*, specific maintenance procedures are listed. The maintenance procedures identified are mandatory for proper use and operation of the *ABX PENTRA 60*. Failure to execute any of these recommended procedures may result in poor reliability of the system.

IMPORTANT

Failure to execute any of these recommended procedures may result in poor reliability of the system.

5.2. Blood specimens

• Verification of any abnormal test result (including flagged results or results outside of the normal range) should be performed using reference methods or other standard laboratory procedures for conclusive verification of the results. The sections below list known limitations of automated blood cell counters which use the principles of impedance and light absorbance as principles of measurement.

5.3. Known interfering substances

WBC, White Blood Cells (Leukocytes)
WBC results that exceed the linearity limits of the system will require dilution of the blood sample (Leukemia sample followed by a leukopenia).
Re-assaying the diluted sample will help to obtain the correct assay value.

Unlysed Red Cells - In some rare instances, the erythrocytes in the blood sample may not be completely lysed. These non-lysed red blood cells may be detected on the WBC histogram with an L1 alarm or as an elevated baseline on the side (leading edge) of the lymphocytes population. Non-lysed erythrocytes will cause a falsely elevated WBC count.

Multiple myeloma - The precipitation of proteins in multiple myeloma patients may give high WBC counts.

Leukemia - A very low WBC count may result from this disease because of possible increased fragility of the leukocytes leading to destruction of some of these cells during counting. These white cell fragments will also interfere with the white cell differential parameters.
These white cell fragments will also interfere with the white cell partial differential parameters: LYM% + #, MON% + #, GRAN% + #. A suspiciously low WBC count may also be seen in patients with lymphocytic leukemias due to the presence of abnormally small lymphocytes which may not be counted by the instrument.

Chemotherapy - Cytotoxic and immunosuppressive drugs may increase the fragility of the leukocytes which may cause low WBC counts.

Cryoglobulins - Increased levels of cryoglobulin that may be associated with myeloma, carcinoma, leukemia, macroglobulinemia, lymphoproliferative disorders, metastic tumors, autoimmune disorders, infections, aneurism, pregnancy, thromboembolic phenomena, diabetes, etc..., which can increase the WBC, RBC or PLT counts and the HGB concentration. The specimen must be warmed up to 37°C (99°F) in a bain marie for 30 minutes and analyzed again immediately after (analyzer or manual method).

Macrothrombocytes - In excessive numbers may affect and increase Leukocyte numeration.

RBC, Red Blood Cells (Erythrocytes)
The red blood cell dilution contains all the formed elements in the blood: erythrocytes, leukocytes and platelets. During erythrocytes counting (red blood cells), platelets are not counted as their size falls below the minimum threshold.

Agglutinated erythrocytes - May cause a low incorrect RBC count. Blood samples containing the agglutinated red blood cells may be suspected by elevated MCH and MCHC values and shown by examination of the stained blood film.

Cold agglutinins - IgM immunoglobulins which are high in cold agglutinin disease may cause lower RBC and PLT counts and increase MCV.

Hgb (Hemoglobin)

Turbidity of the blood sample - Any number of physiological and/or therapeutic factors may produce high incorrect HGB results. To obtain accurate hemoglobin results when increased turbidity of the blood sample occurs, determine the cause of the turbidity and follow the appropriate method below.

High WBC - An extremely high WBC will cause excessive light scatter. In these cases use reference (manual) methods.The diluted sample should be centrifuged, and the supernatant fluid measured with a spectrophotometer.

High lipid concentration - A high concentration of lipids in the blood sample will give the plasma a «milky» appearance. This condition can occur with hyperlipidemia, hyperproteinemia (as in gammapathies) and hyperbilirubinemia. Accurate hemoglobin determinations can be achieved by using reference (manual) methods and a plasma blank.

Increased turbidity may also be seen in cases where the red blood cells are resistant to lysing. This condition will cause an incorrect high HGB result, but may be detected by observing the abnormal MCH, MCHC values, and the increased baseline on the leading edge of the WBC histogram. Erroneous hemoglobin results will cause the results of the MCH and MCHC to be incorrect as well.

Fetal bloods - The mixing of fetal and maternal bloods may produce a high inaccurate HGB value.

Hct (Hematocrit)

Red blood cells agglutination - May produce an inaccurate HCT and MCV values. Red blood cell agglutination may be detected by observing abnormal MCH and MCHC values, as well as by examination of the stained blood film. In such cases, manual methods may be required to obtain an accurate HCT value.

MCV (Mean Corpuscular Volume)

Red blood cells agglutination - May produce an inaccurate MCV value. Red blood cell agglutination may be detected by observing abnormal MCH and MCHC values, as well as by examination of the stained blood film. In such cases, manual methods may be required to obtain an accurate MCV value.

Excessive numbers of large platelets and/or the presence of an excessively high WBC count may interfere with the accurate determination of the MCV value. In such cases, careful examination of the stained blood film may reveal the error.

MCH (Mean Corpuscular Hemoglobin)

The MCH is determined according to HGB value and the RBC count. The limitations listed for the HGB and RBC will have an effect on the MCH and may cause inaccurate values.

MCHC (Mean Corpuscular Hemoglobin Concentration)

The MCHC is determined according to the HGB and HCT values. The limitations listed for the HGB and HCT will have an effect on the MCHC and may cause inaccurate values.

RDW (Red blood cell Distribution Width)
The red blood cell distribution width is determined according to the RBC count.

Nutritional deficiency or blood transfusion - May cause high RDW results due to iron and/or cobalamin and/or folate deficiency.

Plt (Platelets)
WBC fragments may interfere with the proper counting of platelets and cause elevated PLT counts.

Very small erythrocytes (microcytes), erythrocyte fragments (schizocytes).

Agglutinated erythrocytes - May trap platelets, causing an erroneously low platelet count. The presence of agglutinated erythrocytes may be detected by observation of abnormal MCH and MCHC values and by careful examination of the stained blood film.

Hemolysis - Hemolysed specimens contain red cell stroma which may increase platelet counts.

Giant platelets in excessive numbers - May cause a low inaccurate platelet count as these large platelets may exceed the upper threshold for the platelet parameter and are not counted.

Platelet agglutination - Clumped platelets may cause a decreased platelet count and/or a high WBC count. The specimen should be recollected in sodium citrate anticoagulant to ensure the anticoagulated character depending on agglutination and reanalyzed only for the platelet count. The final PLT result must be corrected for the sodium citrate dilution effect. However, these platelet clumps do trigger flags L1, LL and LL1.

A.C.D. blood - Blood anticoagulated with acid-citrate-dextrose may contain clumped platelet which could decrease the platelet count.

Chemotherapy - Cytotoxic and immunosuppressive drugs may increase the fragility of these cells which may cause low PLT counts. Reference (manual) methods may be necessary to obtain an accurate platelet count.

Elevated lipids and/or cholesterol - May interfere with correct platelet counting.

IMPORTANT

Blood samples collected in EDTA will not maintain a stable Mean Platelet Volume. Platelets collected in EDTA swell depending on the time post-collection and storage temperature.

MPV (Mean Platelet Volume)
Giant platelets that exceed the upper threshold of the Platelet parameter may not be counted as platelets. Consequently, these larger platelets will not be included in the instrument's calculation of Mean Platelet Volume.

Very small erythrocytes (microcytes), erythrocytic fragments (Schizocytes) and white blood cell fragments may interfere with the proper counting and sizing of Platelets.

Agglutinated erythrocytes - May trap Platelets, causing an incorrect MPV result. The presence of agglutinated erythrocytes may be detected by observation of abnormal MCH and MCHC values and by careful examination of the stained blood film.

Chemotherapy - May also affect the sizing of PLTs.

LYM# (Lymphocyte count absolute value), LYM% (Lymphocyte percentage)
The Lymphocyte count is derived from the WBC count. The presence of erythroblasts, certain parasites and erythrocytes that are resistant to lysis may interfere with an accurate LYM count. Limitations listed for the WBC count pertain to the LYM# and % counts as well.

MON# (mononuclear cell count absolute), MON% (Mononuclear percentage)
The mononuclear cell count absolute is derived from the WBC count. The presence of large lymphocytes, atypical lymphocytes, blasts and an excessive number of basophils may interfere with an accurate monocyte count. Limitations listed for the WBC count pertain to the MON# and % counts as well.

NEU# (neutrophil count absolute), NEU% (Neutrophil percentage)
The neutrophils cell count is derived from the WBC cell count. The excessive presence of eosinophils, metamyelocytes, myelocytes, promyelocytes, blasts and plasma cells may interfere with an accurate neutrophils count.

EOS# (Eosinophil cell count absolute), EOS% (Eosinophil percentage)
The eosinophil cell count is derived from the WBC cell count. The presence of abnormal granules (degranulated areas, toxic granules...) may interfere with the eosinophil counting.

BAS# (Basophil cell count absolute), BAS% (Basophil percentage)
The Basophil cell count is derived from the WBC cell count.

DESCRIPTION & TECHNOLOGY

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1. PENTRA 60 DESCRIPTION

1.1. Pentra 60 general description

+ Cover
• *Before any attempt to remove the cover, open the pneumatic access door.*

+ Pneumatic access door
• *Allows the operator to access hydraulics parts for maintenance operations. It is mandatory to keep the door locked during the measuring cycles as it ensures the heating of the dilutions.*

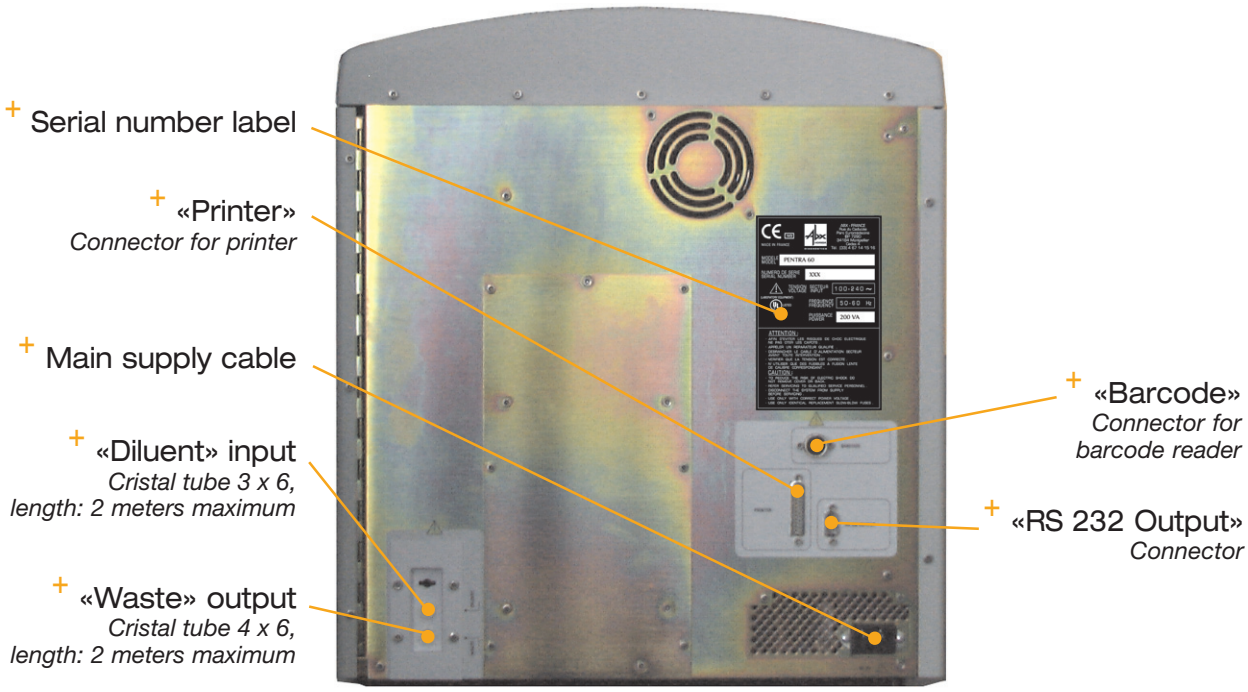
+ Control panel
• *This panel allows the operator to communicate with the instrument:*
- To access to different cycles,
- To identify the patients,
- To setup the instrument, etc...

+ Reagent access door
• *Access to the reagent bottles in order to replace them when empty.*

+ Sampling probe
• *This needle is used for the 53µl whole blood sampling.*

+ LEDs
• *When Green LED is lit instrument is ready to sample,*
• *When Green and Red LEDs are blinking alternatly, instrument is busy sampling,*
• *When Red LED is lit instrument is running.*

1.2. Pentra 60 back side

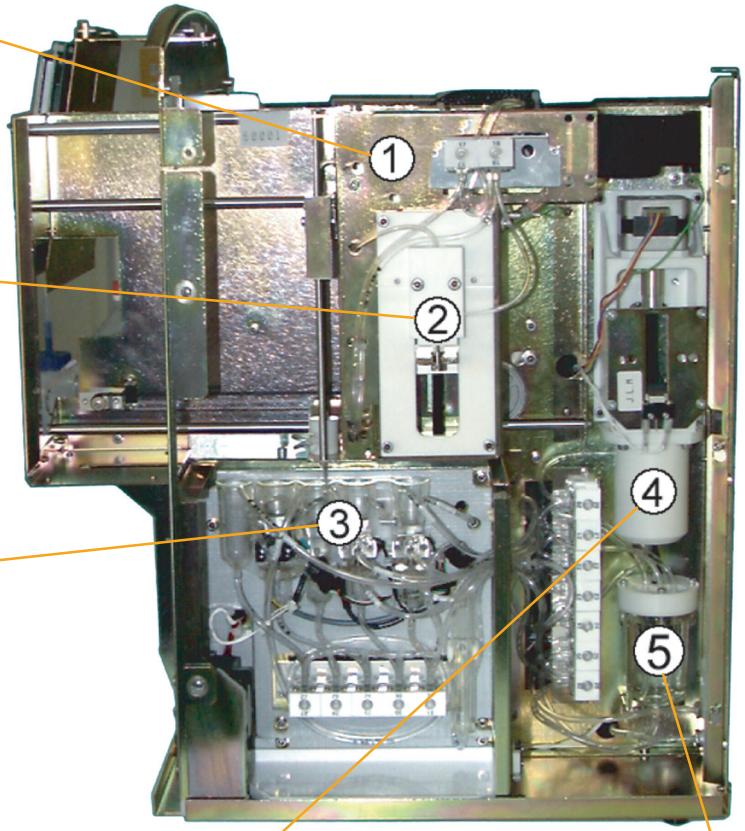


1.3. Mechanical and hydraulic modules

+ Piercing carriage
- *Ensures needle positioning for the different sampling stages and distribution,*
- *Supports the sampling syringe and the blood distribution.*

+ Sampling syringe
- *Distributes portions of the specimen into the dilution chambers,*
- *Takes the sample from the first dilution and distributes it into the RBC/PLT chamber.*

+ Count assembly
- *Receives the different rinsings and dilutions,*
- *Regulates the temperature of dilutions,*
- *Provides the dilutions for WBC/BASOS, RBC/Plt and Hgb.*



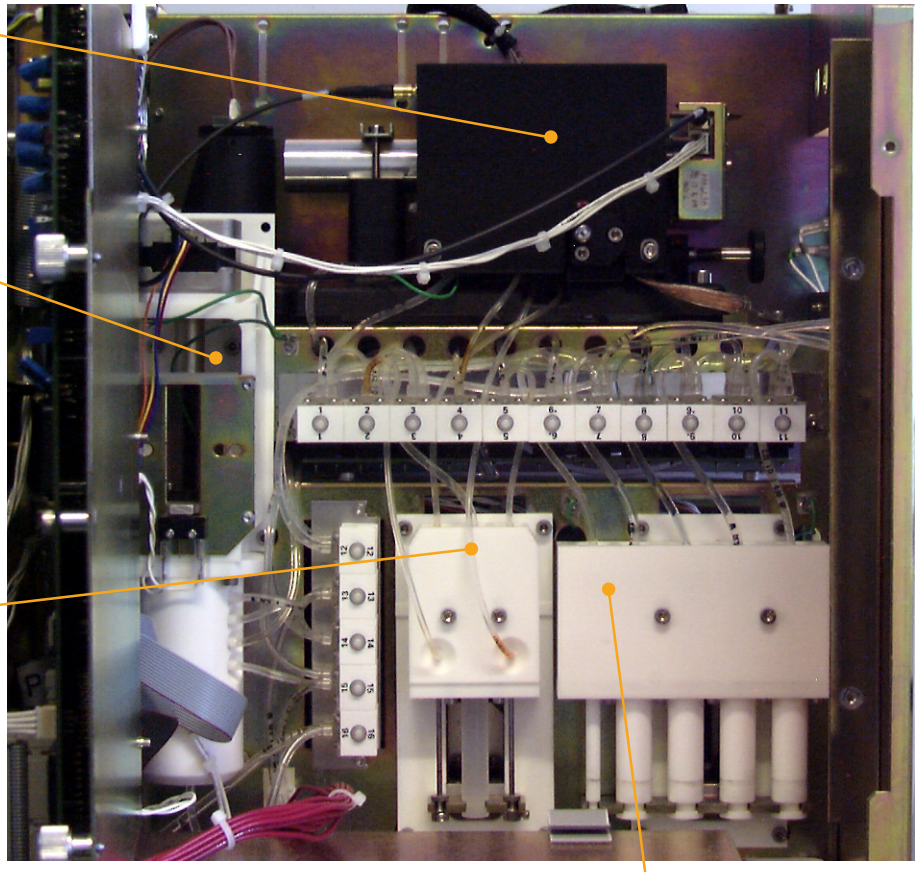
+ Draining syringe
- *Drains the chambers,*
- *Bubbles the mixtures,*
- *Transfers, by vacuum, the LMNE specimen from the mixing chamber towards to the injector of the optical flowcell.*

+ Diluent tank
- *Contains the necessary diluent for an analysis cycle,*
- *Prevents Diluent degassing as it is being aspirated by the syringes,*
- *Is vacuum filled by the counting syringe.*

+ Optical bench
• *Ensures the support and adjustment of the flowcell, lamp, and optical and electronic elements.*

+ Counting syringe
• *Ensures the vacuum for the WBC and BASOS counts,*
• *Ensures the vacuum for the RBC and the Plts counts,*
• *Ensures the vacuum for filling the diluent tank with diluent.*

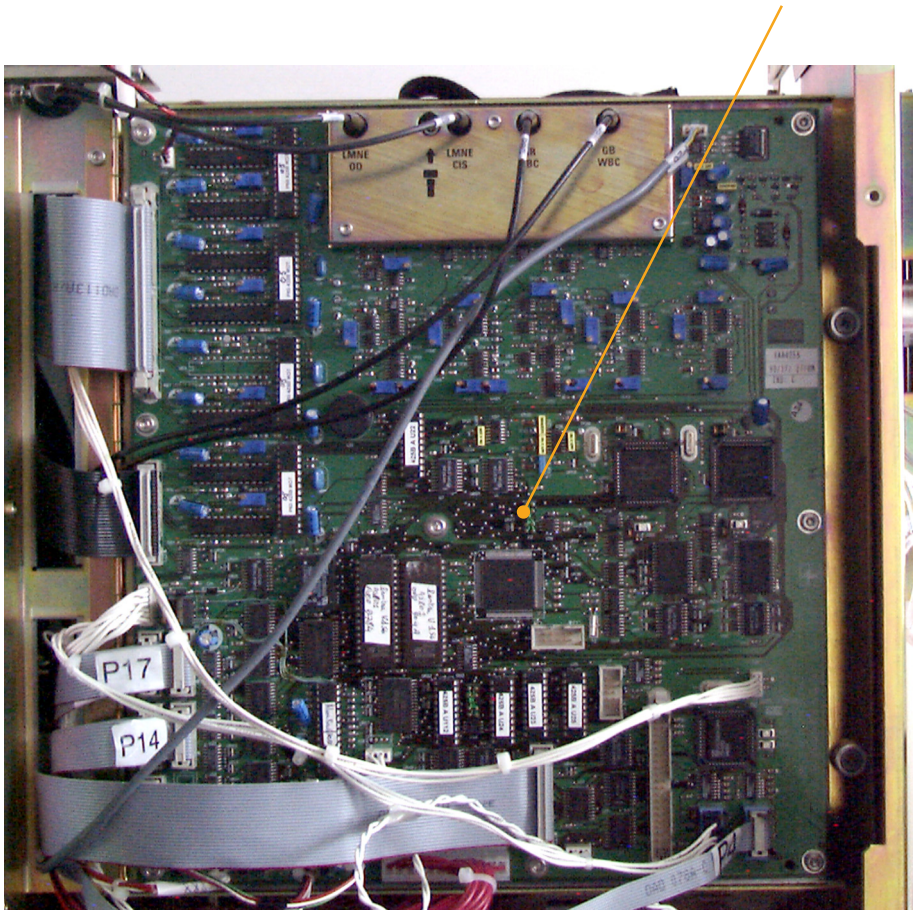
+ LMNE Syringe assembly
• *Ensures the correct proportioning of the stop diluent in the LMNE chamber,*
• *Injects the specimen into the flowcell,*
• *Injects the interior and exterior sheath into the flowcell.*



+ Reagent Syringe assembly
• *Ensures the correct proportioning of the reagents:*
• *Lysing reagent for hemoglobin (ABX **ALPHALYSE**),*
• *Cleaning reagent (ABX **CLEANER**),*
• *Lysing reagent for DIFF count (ABX **EOSINOFIX**),*
• *Diluent used for the dilutions (ABX **DILUENT**) except the LMNE stop diluent,*
• *Lysing reagent for WBC/BASO (ABX **BASOLYSE II**).*

+ Main board

Located on the left side of the instrument. The board is fastened onto a door in order to allow access to the fluidic modules.



Main functions of the board

- *Amplifies, processes and counts the following signals:*
  - *Resistive signals and LMNE optical signals,*
  - *RBC signal,*
  - *Plt signal,*
  - *WBC/BASO signals.*
- *Measures hemoglobin.*
- *Pilots the motorised elements.*

CAUTION

When opening the main board support panel, be careful not to disconnect or damage electric cables.

2. MEASURING PRINCIPLES

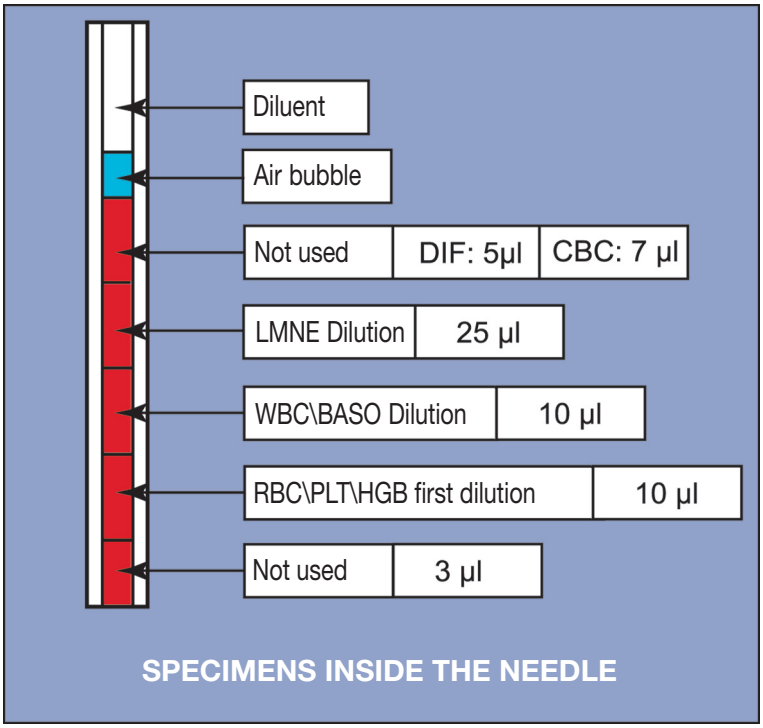
2.1. Multi Distribution Sampling System (MDSS)

+ *ABX PENTRA 60 analysis cycle needs 3 blood specimens distributed as follows:*

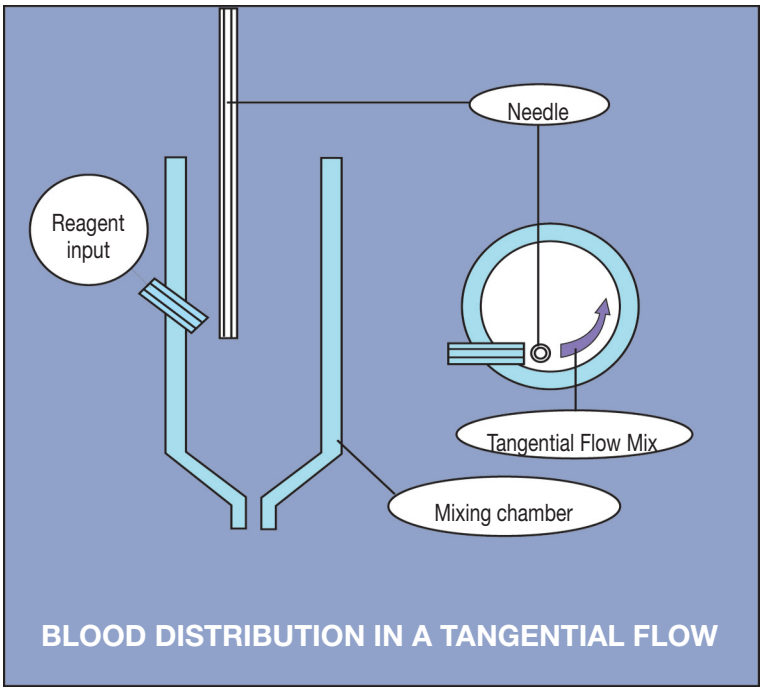
- *one specimen for the first RBC/Plt dilution and the Hgb measurement.*
- *another specimen for the BASO/ WBC count.*
- *the last specimen for the LMNE matrix.*

***These 3 specimens** are provided by means of a «multi distribution» principle:*

- *53 µl of blood are aspirated inside the needle (sufficient volume for all dilutions) then divided up and distributed to the chambers with reagents.*
- *Extremities of the specimen are not delivered to the dilutions.*



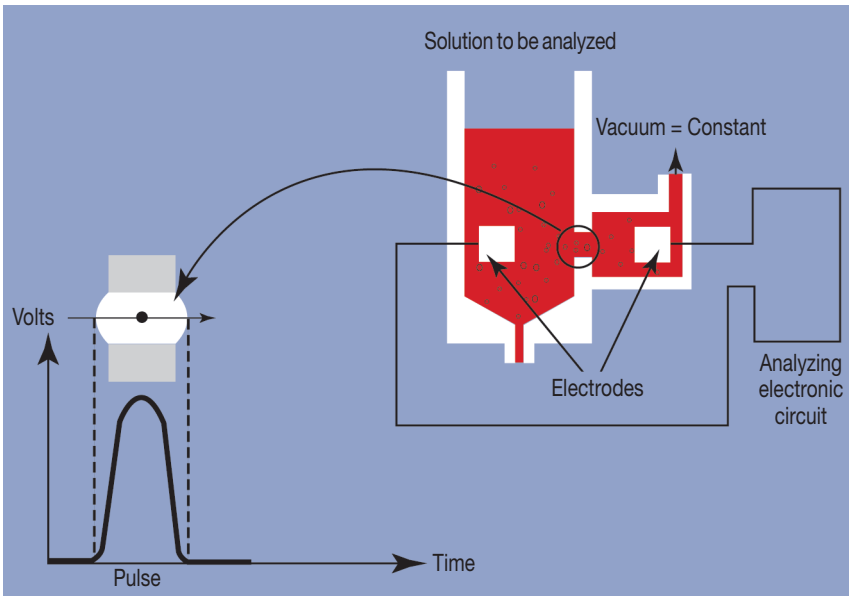
+ ***Specimen distribution** in the chambers is carried out in a tangential flow of reagent which allows perfect mixing of the dilution and avoids any viscosity problems (this multi distribution in a reagent flow is an HORIBA ABX patent).*



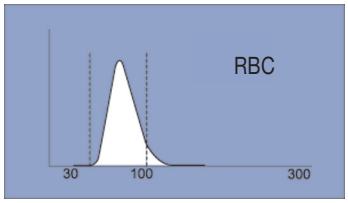
2.2. RBC / Plt detection principles

Measurement of impedance variation generated by the passage of cells through a calibrated microaperture.

- *The specimen is diluted in an electrolytic diluent (current conductor) and pulled through the calibrated microaperture. Two electrodes are placed on either side of the aperture. Electric current passes through the electrodes continuously.*
- *When the cell passes through the aperture, electric resistance between the two electrodes increases proportionately with the cell volume.*
- *The generated impulses have a very low voltage, which the amplification circuit increases, so that the electronic system can analyze them and eliminate the background noise.*

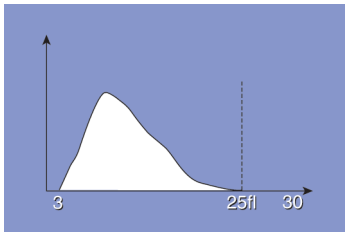


TECHNICAL CHARACTERISTICS OF THE RED BLOOD CELL AND PLATELET COUNTS			
Initial blood volume	10 µl	Method	Impedance
Vol. ABX DILUENT	2500 µl	Aperture diameter	50 µm
Final dilution rate**	1/10000	Count vacuum	200 mb
Temperature of reaction	35°C	Count period	2 X 5 seconds
**: TWO SUCCESSIVE DILUTIONS ARE CARRIED OUT : PRIMARY DILUTION FOR RBC AND PLT:			
Blood (µl): 10			
ABX DILUENT (µL):	1700	dilution:	1/170
SECONDARY DILUTION RBC AND PLT (FROM THE PRIMARY DILUTION):			
Dilution (µl): 42.5			
ABX DILUENT(µL):	2500	dilution:	1/58.8
FINAL DILUTION: 1/170 X 1/58.8 =1/10000			



Results

• Number of cells counted per volume unit x Calibration coefficient.



Histograms

RBC: Distribution curves on 256 counting channels from 30fl to 300fl.

Plt: Distribution curves on 256 channels from 2fl to a mobile threshold. This threshold moves according to the microcyte population present in the analysis area.

2.3. Hgb measurement

During the analysis cycle, lysing reagent is released into the Dilution chamber.

• **Alphalyse**

This reagent breaks down the RBC cell membrane and releases the hemoglobin within the cell. The hemoglobin, released by the lysing reagent, combines with the potassium cyanide from the lysing reagent to form a chromogenous cyanmethemoglobin compound. This compound is then measured through the optical part of the first dilution chamber by way of spectrophotometry at a wavelenght of 550 nm.

• **Lysebio**

Reagent for erythrocyte lysis and cyanide-free determination of hemoglobin. All the heme iron is oxidized and stabilized producing chromogenic species for quantitation by spectrophotometry at a wavelength of 550 nm.

TECHNICAL CHARACTERISTICS FOR THE MEASUREMENT OF THE HEMOGLOBIN:			
Volume of blood	10 µl	Method	Photometry
Volume ABX DILUENT	1700 µl	Wavelength	550 nm
Volume LYSE	400 µl		
Complement ABX DILUENT	400 µl		
Final dilution rate	1/250		
Temperature of reaction	35°C		

Results

• Hemoglobin results are given as such: Absorbance value obtained from the sample x Coefficient of calibration.

2.4. Hct measurement

• The height of the impulse generated by the passage of a cell through the microaperture is directly proportional to the volume of the analyzed RBC.

• The hematocrit is measured as a function of the numeric integration of the MCV.

2.5. RDW calculation

• The study of the RBC distribution detects erythrocyte anomalies linked to anisocytosis.
A Red Cell Distribution Width (RDW) will enable you to follow the evolution of the width of the curve in relation to the cell number and average volume.

$$RDW = \frac{K\ SD}{MCV}$$

With:
• K = system constant.
• SD = determined Standard Deviation according to statistical studies on cell distribution.
• MCV = Mean Corpuscular Volume of erythrocytes.

2.6. MCV, MCH, MCHC calculation

• MCV (Mean Cell Volume) is calculated directly from the RBC histogram.

• MCH (Mean Cell Hemoglobin) is calculated from the Hgb value and the RBC number.
The mean hemoglobin weight in each RBC is given by the formula:

$$MCH\ (pg) = \frac{Hgb}{RBC} x\ 10$$

• MCHC (Mean Corpuscular Hemoglobin Contained) is calculated according to the Hgb and Hct values. Mean Hgb concentration in the total volume of RBC is given by the formula:

$$MCHC\ (g/dL) = \frac{Hgb}{Hct}\ x\ 100$$

2.7. MPV Measurement

• The MPV (Mean Platelet Volume) is directly derived from the analysis of the platelet distribution curve.

2.8. Pct calculation

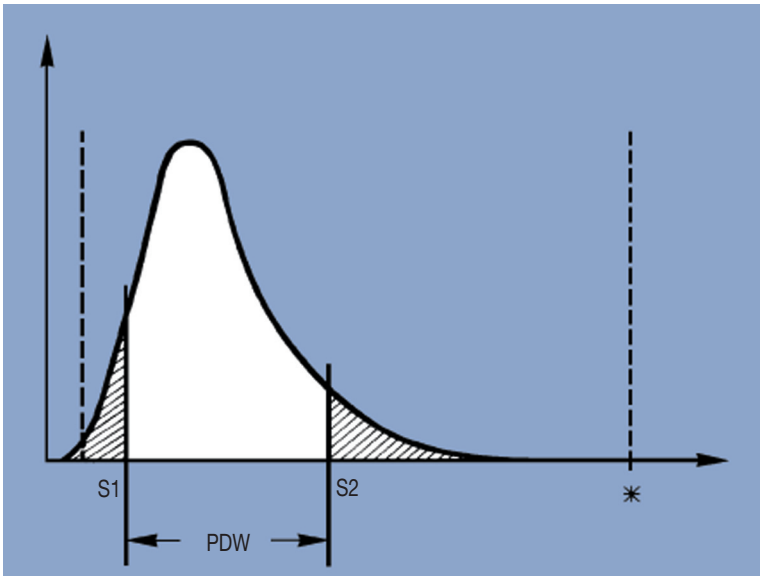
• Thrombocrit is calculated according to the formula:

$$Pct\% = \frac{Plt\ (10^3/\mu l) \times MPV\ (\mu m^3)}{10\ 000}$$

2.9. PDW calculation

• PDW (Platelet Distribution Width) is calculated from the Plt histogram.

• The PDW is represented by the width of the curve between 15% of the number of platelets starting from 2fl (S1), and 15% of the number of platelets beginning with the variable top threshold (S2).



2.10. WBC and differential count

2.10.1. General principles

The WBC count is carried out twice by two different sensors:
- In the BASO count chamber at the same time as the BASOS count,
- In the optical chamber during the acquisition of the LMNE matrix.

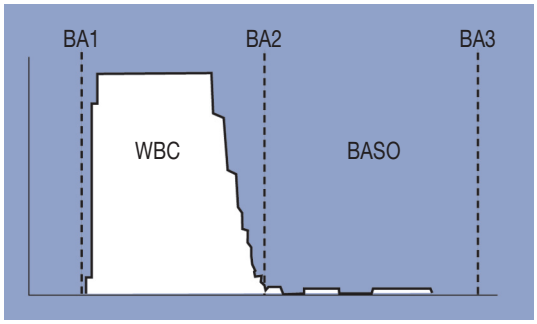
The reference count is the one obtained in the WBC and BASO count chamber.

2.10.2. BASO / WBC Count

- Detection principle is the same as for RBC.

Differentiation betwen BASOs and other leukocytes is obtained by means of the **BASOLYSE II** specific lysing action.

- All the WBCs are counted between the electrical threshold <**0**> threshold <**BA3**>. The basophils are located from threshold <**BA2**> to threshold <**BA3**>.



Technical Characteristics of the WBC/BASO count			
Blood volume	10 µl	Method	Impedance
BASOLYSE II volume	2000 µl	Ruby diameter	80 µm
Dilution rate	1/200	Depression of count	200 mb
Reaction temperature	35°C	Duration of the count	2 X 6 seconds

Results

WBC: Number of cells per volume x coefficient of calibration.

BASO: Number of cells per volume x coefficient of calibration in percentage regarding the total number of leukocytes (BASO + WBC nuclei).

2.10.3. LMNE matrix

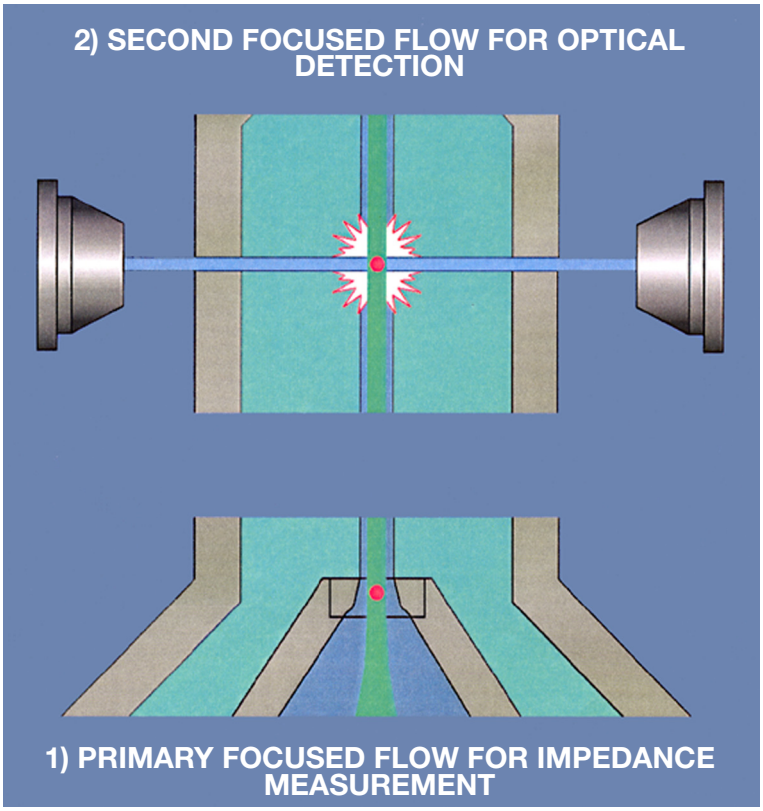
• *The WBC and Differential count are based on 3 essential principles:*

1 - The double hydrodynamic sleeving
«DHSS» (HORIBA ABX patent).

2 - The volume measurement
impedance changes.

3 - The measurement of transmitted light
with 0° angle, which permits a response according to the internal structure of each element and its absorbance by means of incident light diffusion.

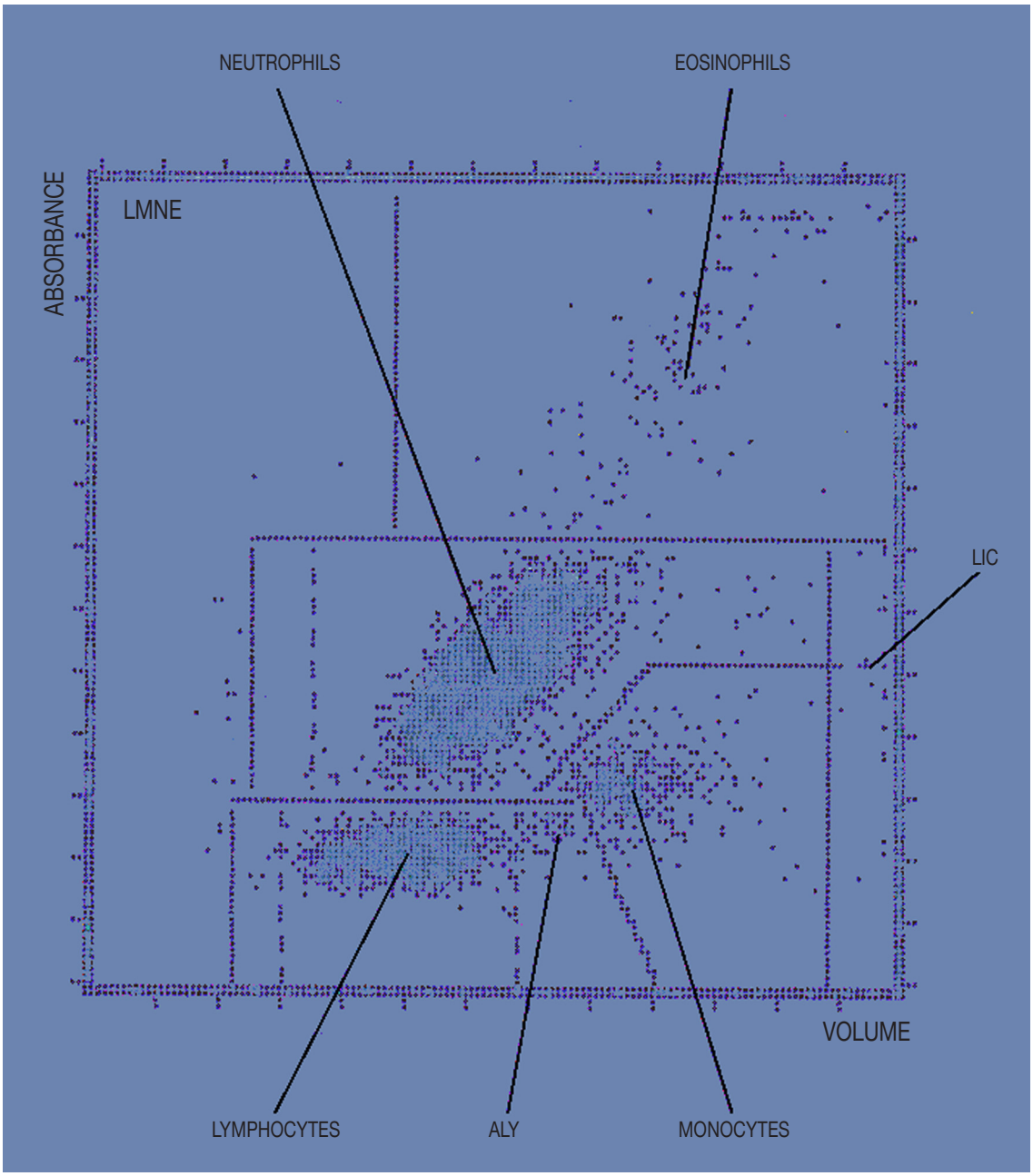
• *25µl of whole blood is delivered to the LMNE chamber in a flow of EOSINOFIX. This reagent lyses the RBC, stabilizes the WBC in their native forms and stains the eosinophil nuclei with a specific coloration.*
• *The solution is then stabilized with diluent and transferred to the measuring chamber. Each cell is measured both in absorbance (cytochemistry) and resistivity (volume).*



TECHNICAL CHARACTERISTICS OF THE WBC COUNT DURING THE ACQUISITION OF THE MATRIX			
Blood volume	25 µl	Method	Impedance with hydrofocus
Eosinofix volume	1000 µl	Ruby diameter	60 µm
Diluent volume	1000 µl	Flow diameter	42 µm
Final dilution rate	1/80	Injection duration	12 seconds
Reaction temperature	35°C	Volume injected	72 µl
Incubation duration	12 s.		

Results

• From these measurements, a matrix is drawn up with volumes on the X-axis and optical transmission on the Y-axis.
The study of the matrix image permits the clear differentiation of 4 out of 5 leukocyte populations. As a matter of fact, the basophil population is very small compared to the other 5 in a small blood sample.


NEUTROPHILS
EOSINOPHILS
ABSORBANCE
LMNE
LIC
VOLUME
LYMPHOCYTES
ALY
MONOCYTES

***MONOCYTES**: The monocytes, being cells with large kidney shaped nuclei and a large non-granular cytoplasm, will neither be scattered nor absorb a large amount of light. They will therefore be positioned in the lower part of the optical axis but clearly to the right of the volume axis. Certain large monocytes can be found on the right side of the matrix in the lower LIC (Large Immature Cells) zone. The immature granulocytic cells are detected by their larger volumes and by the presence of granules which increase the intensity of the scattered light. Therefore, cells such as metamyelocytes will be found clearly to the right of the neutrophils and nearly at the same level. Myelocytes and promyelocytes will be found in saturation position on the far right of the matrix. These last three populations will be counted as LIC (Large Immature Cells) and their given results are included in the neutrophil value. The blast cells will be found generally to the right of the monocytes, and, as such, will increase the LIC count. Small blasts will be found between the normal lymphocytes and monocytes. Platelets and debris from erythrocyte lysis represent the background noise population located in the lower left area of the matrix. Most of the population partition thresholds are fixed and give the limits of the morphological normality of leukocytes. Changes in the morphology of a population will be expressed on the matrix by a shifting of the corresponding population.*

***LYMPHOCYTES**: The lymphocytes being small with regular shape, are positioned in the lower part of both the optical axis and volume axis. Normal lymphocyte populations are generally observed with a good volume homogeneity. Large lymphocytes are detected in the ALY (Atypical Lymphocytes) zone, where reactive lymphoid forms, stimulated lymphocytes and plasmocytes are also to be found. The far left side of the lymphocyte zone should normally be empty, but when small lymphocytes are present, population may exist in this area. The presence of platelet aggregates is detected by a distribution pattern that moves from the origin of the matrix (background zone) into the lymphocyte zone. The NRBCs with their cytoplasmic membranes lysed like the erythrocytes, will have their nuclei situated to the far left side of the lymphocyte zone.*

***EOSINOPHILS**: With reagent action on cytoplasmic membranes, the leukocytes keep their native size and only eosinophils are colored for optical separation. Eosinophils will be situated in the upper part of the optical Y-axis due to their strong absorbance qualities and their size, which is nearly equivalent to large neutrophils.*

***NEUTROPHILS**: The neutrophils, with their cytoplasmic granules and their generally segmented nuclei, will scatter light depending on their morphological complexity. A hypersegmented neutrophil will give an increased optical response with respect to a young neutrophil population which will be in the upper position of the optical axis depending on the presence of segmentation and/or granules.*

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1. INSTRUMENT START UP

1.1. Waste levels

- At the beginning of each day check if the waste container needs to be emptied. Be careful Wastes must be handled according to your local/national regulations.

- See *Chapter 5. Maintenance & Troubleshooting* to empty the waste container.

1.2. Printer start up

- Check the printer paper. If more printer paper is needed, replace the printer paper according to the *Printer user manual* supplied with the instrument.

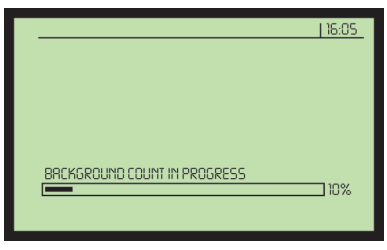
- Press the ON/OFF switch. Check the control LEDs are ON.

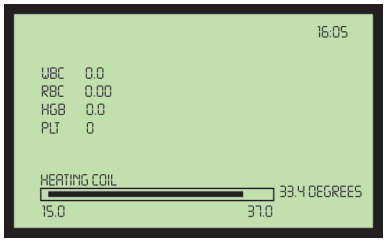
- See *Chapter 4. Instrument configuration* to setup printing mode.

1.3. Pentra 60 start up

- Press the ON/OFF switch located on the left side of the instrument.

- A STARTUP cycle is run automatically:
- A rinse cyle is carried out.
- Then a background count (an analysis cycle on reagent without any blood specimen).





• If the background counts are not within acceptable limits the message «**STARTUP FAILED**» is displayed.

Background count limits:
WBC = 0.3 x10^3/mm^3
RBC = 0.03 x10^6/mm^3
Hgb = 0.3 g/dl
Plt = 7.0 x10^3/mm^3

IMPORTANT

The instrument will not operate if the reagent temperature is under 35°C (69°F). If required a bargraph is displayed after start up to check and wait for temperature progression.

• If a low reagent level is expected during the day, the message «**REAGENT LOW LEVEL**» is displayed.
Proceed as described in *Chapter 5. Maintenance & Troubleshooting*.

WARNING

It is mandatory to power down the system if not used more than a 36 hour period. This eliminates the possibility of the dilution chambers evaporating, causing startup alarm.

2. SPECIMEN COLLECTION AND MIXING

• All blood samples should be collected using proper technique.

Consider all Specimens, Reagents, Calibrators, Controls, etc… that contain human blood or serum as potentially infectious! Use established, good laboratory working practices when handling specimens. Wear protective gear, Gloves, Lab coats, Safety glasses and/or Face shields, and follow other bio-safety practices as specified in OSHA Blood borne Pathogens Rule (29 CFR part 1910. 1030) or equivalent biosafety procedures.

• When collecting blood specimens, Venous blood is recommended, but Arterial blood may also be used in extreme cases. Blood collection must be placed in a Vacuum or atmospheric collection tubes.
For additional information on collecting venous and capillary blood samples, refer to NCCLS document H3-A4 and NCCLS document H4-A4 (sept 1999).

• The sample collection tube has to be filled to the exact quantity of blood indicated on the tube itself. Any incorrectly measured blood sample collections will show a possible variation in the results.

2.1. Recommended anticoagulant

• The recommended anticoagulant is K3EDTA with the proper proportion of blood to anticoagulant as specified by the tube manufacturer.
K2EDTA is an acceptable alternative, as long as the sample collection is made in normal conditions.
Otherwise, blood clots may be possible.

2.2. Blood sample stability

• Specimens may be used between 15/20 minutes after collection. The results on all parameters depend on the mode of conservation of the sample.
Depending on the parameter to be measured, the sample stability may be up to 48 hours.

2.3. Microsampling

• The «Open tube» sampling mode enables the user to work with 100µl microsamples (for pediatrics and geriatrics).

2.4. Mixing

• The blood samples must be gently and thoroughly mixed just before placing them into the tube holder and closing the tube holder door. This will ensure a homogeneous mixture for measurement.

3. CALIBRATION VERIFICATION (CONTROL BLOOD SAMPLING)

CAUTION

The calibration on HORIBA ABX instruments is an exceptional procedure, which must be carried out particularly in case of certain technical interventions (installation, maintenance, service intervention).
The calibration should not be carried out to compensate a drift on a result due for example to a clogging of the instrument.

• Before carrying out a calibration, it is essential to make sure that the instrument is in perfect condition of operation, and to follow the steps below:

1- Carry out a concentrated cleaning (refer to the corresponding paragraph of the manual).

2- Perform two blank cycles to check the cleanliness of the instrument (if the blank measurement is not correct, please contact your *HORIBA ABX* Representative).

3- Check the repeatability of the instrument by running six times a normal human blood without taking account of the first result (if the repeatability is not correct, please contact your *HORIBA ABX* Representative).

4- Calculate the CV obtained out on the 5 results, the values of CV must be lower than those given in the manual.

5- Run a control blood and check that the values are within the acceptable limits. If not, run a new control.

• If the instrument is clean (blank cycles in conformity with the values given in the manual), that repeatability is correct (values of CV acceptable) and that the values of control are not within the acceptable limits, then it is possible to carry out a calibration:

6- Run at least 4 times Calibrator without taking the values of the first result into account.

7- Calibrate the instrument on the average of the last three results according to indications of the manual. Take care to respect the minimum and maximum calibration coefficient values given in the manual. Run 3 times again Calibrator to check the values.

8- Confirm the calibration while passing a blood of control, the values have to be returned within acceptable limits.

9- After about thirty analyses of the day, check that values of MCV, MCH and MCHC are in conformity with the usual values of the laboratory.

4. RUNNING SPECIMENS

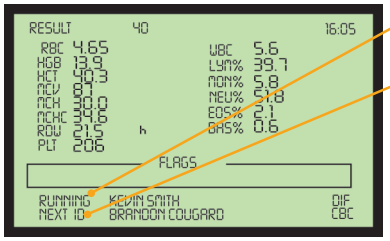
• Two Identification modes are available (see *Chapter 4. Instrume'nt configuration* to configure the ID mode):
1 - alphanumerical mode: requires the patient (or control) identification on each analysis.
2 - Sequence # mode: increments a sequence number on each analysis.

4.1. Alphanumerical mode

• Identification can be entered using the ↓ and ↑ (16 characters, letters or numbers) keys of the front panel.

• Press ← or → for each letter to step to the next one.

• Press Enter when the identification is correct.

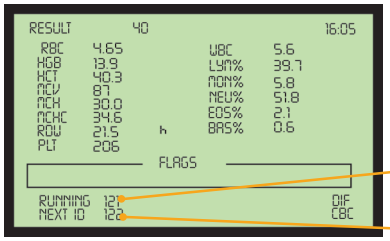


- The identification of the current analysis is shown on the «*RUNNING*» field.
- The identification of the analysis to come is shown on the «*NEXT ID*» field.

NOTE

If no identification has been entered, the analysis cycle will not start.

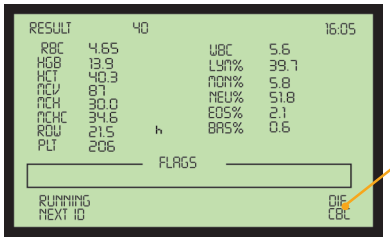
4.2. Sequence # mode



• **Sequence #** (from 1 to 99999) can be entered using the numeric keys of the front panel. Press Enter to record the number.

- The sequence # of the current analysis is shown on the «*RUNNING*» field.
- The sequence # of the analysis to come is shown on the «*NEXT ID*» field.

4.3. Analysis mode selection

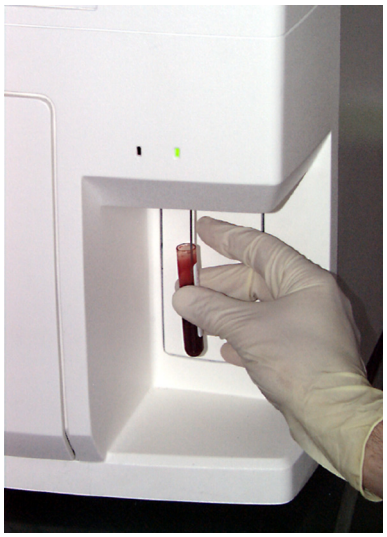


• Press CBC/DIFF to perform the selection between a CBC analysis or a DIF analysis:

- The mode on the current analysis is indicated on the right side of the screen as well as the mode on the analysis to come.

4.4. Analysis

• Present the specimen as shown on the beside diagram and press the sampling bar located behind the sampling needle or press the Start key.



• When the light indicator stops flashing, the LED turns to red, remove the tube.

• When the LED turns to green again, the instrument is ready for the next analysis.

4.5. Automatic cleaning

• When the instrument has run 75 specimens from the date changing, an automatic cleaning procedure is carried out.

• The automatic cleaning frequency can be adjusted by the user from 1 to 75 as described *Chapter 4. Instrument configuration*.

4.6. End of the day rinsing

• It is necessary to run a standby/shutdown cycle at the end of the day.

• Press the Shut down key, the instrument performs a complete cleaning with *ABX **CLEANER***, and puts the system into the standby mode.

• The instrument can be switched off if the working day is completed or left in this standby mode overnight or until the next analysis.

NOTE

A startup cycle is required systematically after a shutdown cycle if the instrument has to be operated.

WARNING

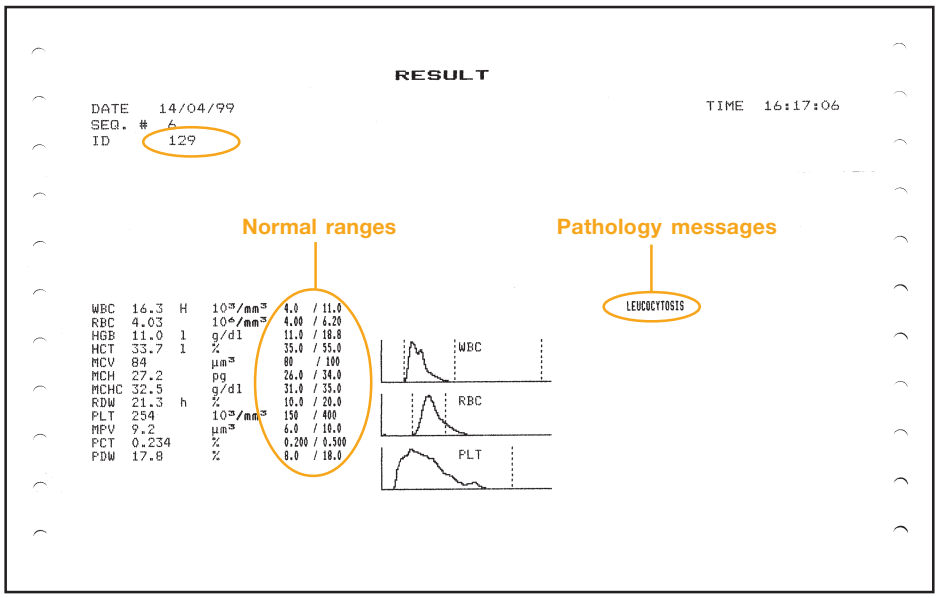
It is mandatory to power down the system if not used more than a 36 hour period. This eliminates the possibility of the dilution chambers evaporating, causing startup alarm.

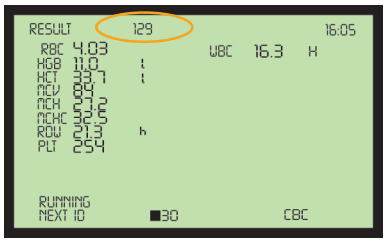
5. RESULTS

• When the analysis cycle is completed, results are displayed and printed out according to the setup of the instrument *Chapter 4. Instrument configuration*:
- Printout configuration («*PRINTER*»)
- Sequence # mode or Alphanumerical mode («*IDENTIFICATION MODE*»)
- Unit setup («UNITS»)
- Possible flags («LAB LIMITS»).

5.1. CBC Mode

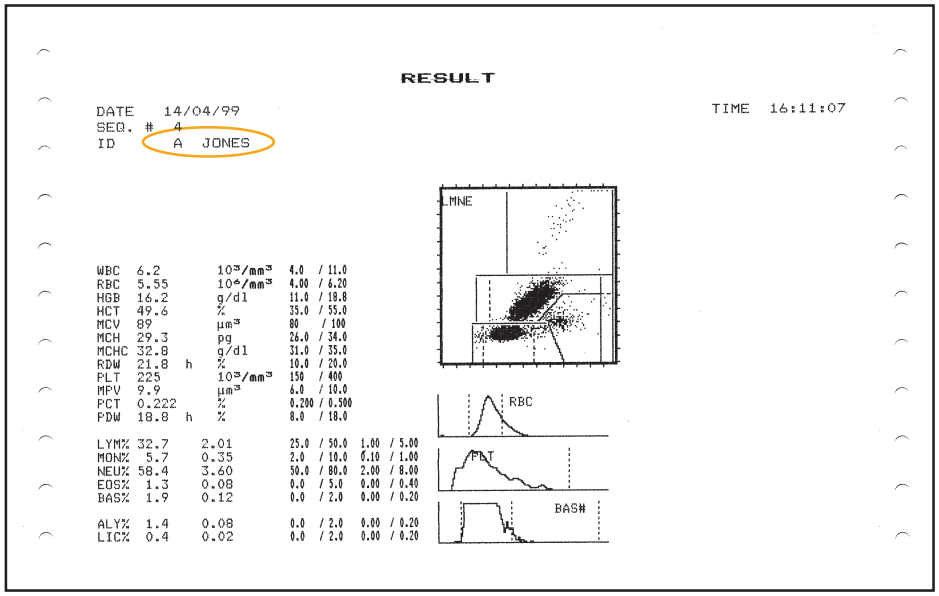
• IDENTIFICATION MODE: Sequence #

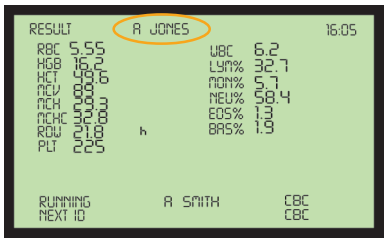




5.2. DIF Mode

- IDENTIFICATION MODE: Alphanumerical mode





5.3. Flags

These flags can be divided up into 5 different groups:
- Flags linked to a result when it exceeds the normality limits.
- Flags linked to a problem in the morphology of blood cell population.
- Flags linked to a result or to instrument operation leading to a «default analysis».
- Flags linked to instrument in use.

NOTE

Each flag sensitivity can be adjusted by the operator (see Chapter 4. Instrument configuration).

5.3.1. Normal and panic ranges

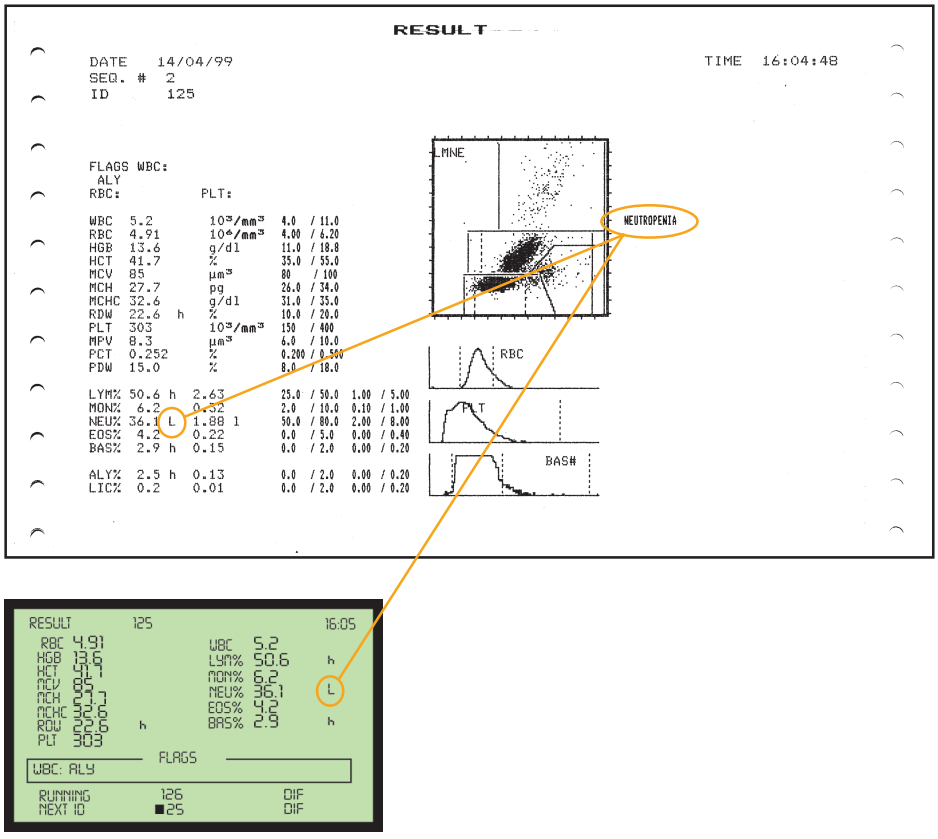
«***h***» indicates that the result is above the normal limit set by the user.

«***l***» indicates that the result is below the normal limit set by the user.

«***H***» indicates that the result is above the panic limit set by the user.

«***L***» indicates that the result is below the panic limit set by the user.

- These flags may be criteria for the pathology messages.



Hgb blank

+ See *Chapter 4. Instrument configuration* to defined BHB# and MHB %.

• At the end of the Startup cycle, the Hgb blank value is controlled. If this value is not within acceptable limits, a reject flag (shown by *****) is triggered on the Hgb parameter.

• On each analysis cycle, the instrument performs a Hgb blank on diluent and checks this measured against the Hgb reference value.
If this Hgb blank value is too different from the mean of the reference values of previous analyzes (higher than BHB# defined by the user) the instrument triggers a suspiscion flag (shown by **!**) on the Hgb parameter.

• (**!**) is also associated with Hgb result if the difference between 3 successive measures on the same sample is higher than MHB % limit defined by the user.

• Sample has to be rerun.

NOTE

No result will be given on Hgb [(----) instead] when 3 suspicion flags (!) associated with the Hgb reference value have been triggered off (MCHC and MCH are also not reported).

Reject (between two counts)

• A reject flag (shown by *) occurs when two counts on a parameter differ more than the pre-defined limits. It indicates that the result is not coherent and the sample has to be rerun.

• ***RBC***

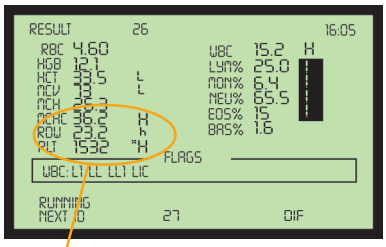
A reject on RBC gives a default analysis value, shown by a * on RBC, MCV, MCH, MCHC and RDW.

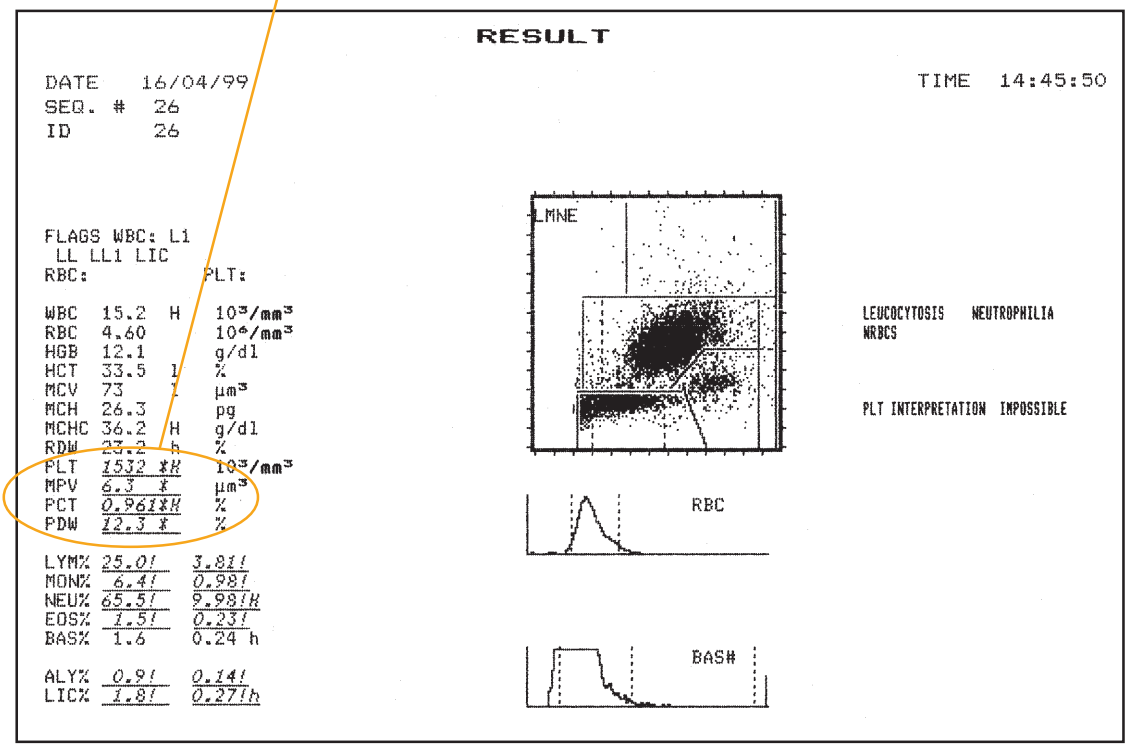
• ***Plt***

A reject on Plt gives a default analysis value, shown by a * on Plt, Pct, MPV and PDW.

• ***WBC***

A reject on WBC gives a default analysis value, shown by a * on WBC and Diff # results.





5.3.2. LMNE matrix flags

Suspiscion

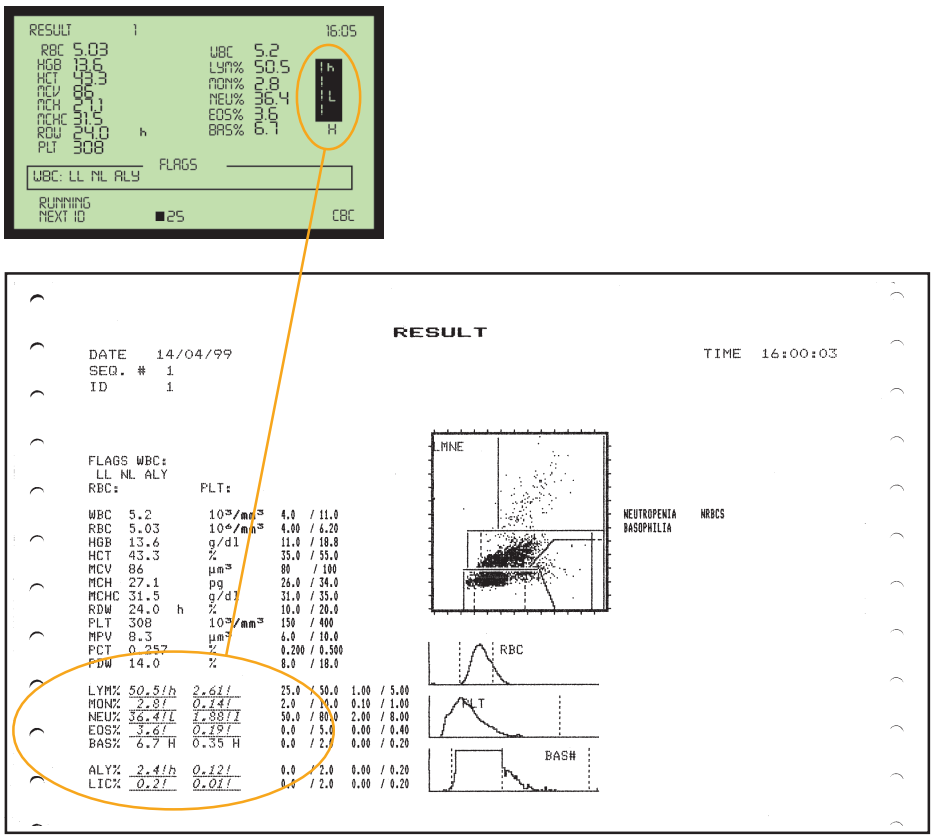
• When populations are detected in abnormal quantity in one or several boxes of the matrix, some flags may occur associated with (**!**).
If one result appears with one or several parameters associated with this (**!**), the result should be further investigated (pathology suspiscion, clotted sample, plasma cells...).

• Twelve different flags may occur regarding the shifting of leukocyte populations on the matrix channel or the presence of abnormal populations.
These flags are: **Reject, NO, LL, LL1, NL**, **MN, RM, LN, RN, NE, ALY, GCI** (see flag signification further in this *Chapter*).

Reject (on LMNE matrix)

• A reject on the LMNE channel indicates a poor correlation between the resistive and the optical measurements on the matrix.
It is shown by a (*) on all the differential parameters in % and #.

• The result is not reliable and specimen must be rerun.



NO flag

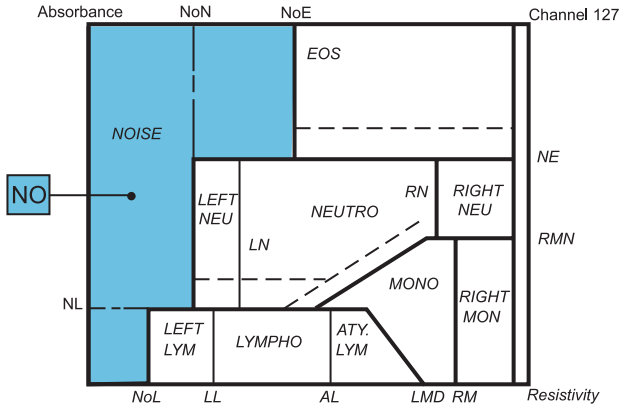
Meaning: Background **NO**ise.

This flag occurs when the number of particles counted in the background noise area is higher than the limit set up in **NO#** or when the number of counted particles versus the total number of WBC, is above the **NO%** limit.

Suspected abnormalities:
- Platelet aggregates,
- Large number of platelets,
- Erythrocyte membrane resistant to lysis (stroma),
- NRBCs,
- Pollution.

+ ***Standard** values for **NO**:*
% 100
 # 120

Adjustment:
see Chapter 4. Instrument configuration.



LL flag

Meaning: **L**eft **L**ymphocytes

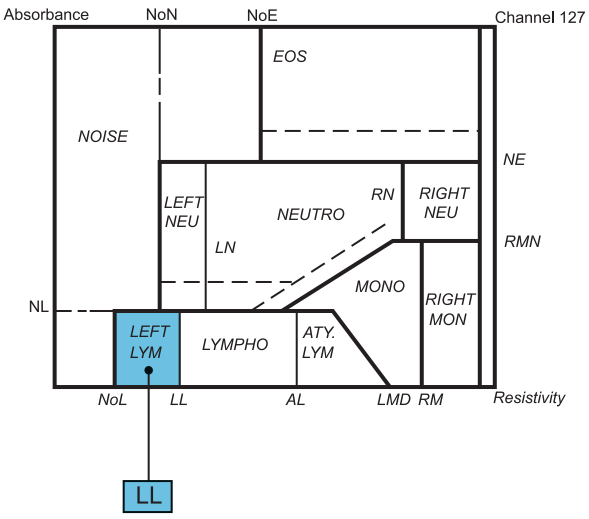
Presence of a significantly large population on the left-hand side of the lymphocyte area. This flag appears when the number of particles counted is higher than the limit set up in **LL#** or when the number of counted particles versus the total number of WBC exceeds the **LL%** limit.

Suspected abnormalities:
- Small lymphocytes,
- Platelets aggregates,
- NRBCs,
- Erythrocyte membrane resistant to lysis (stroma).

This flag occurs associated with an (**!**) on:
- LYM % LYM #
- NEU % NEU #
- MON % MON #
- EOS % EOS #
- ALY % ALY #
- LIC % LIC #

+ ***Standard** values for **LL**:*
% 100
 # 50

Adjustment:
see Chapter 4. Instrument configuration.



LL1 flag

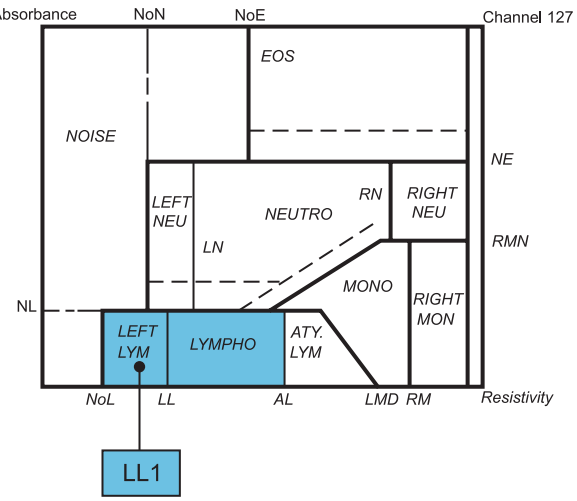
Meaning: **L**eft **L**ymphocytes **1**

Presence of a significantly large population of cells on the left-hand side of the lymphocytes area. This flag occurs when the number of particles counted is higher than the limit set up in **LL1# *and*** when the number of particles counted in **LL** regarding to the total number of lymphocytes is above the **LL1%** limit.

Suspected abnormalities:
- Platelet aggregates,
- NRBCs,
- Erythrocyte membrane resistant to lysis (stroma),
- Stroma,
- Small abnormal lymphocytes.

+ ***Standard** values for **LL1**:*
% 5
 # 45

Adjustment:
see Chapter 4. Instrument configuration.



NL flag

Meaning: **N**eutro/**L**ympho

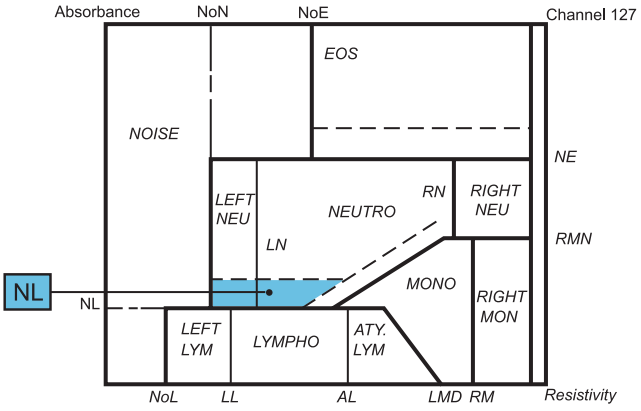
Presence of a significantly large population of cells located in the separation threshold area between lymphocytes and neutrophils. This flag occurs when the number of particles counted in this area is higher than the limit set up in **NL#**, or when the number of counted particles regarding to the total number of WBC is above **NL%** limit.

Suspected abnormalities:
- Small neutrophils without granules and/or slightly segmented,
- Lymphocytes with a segmented nucleus or Activated Lymphocytes,
- Neutrophils with membrane weakness.

This flag occurs associated with an (**!**) on:
- LYM % LYM #
- NEU % NEU #

+ ***Standard** values for **NL**:*
% 3
 # 120

Adjustment:
see Chapter 4. Instrument configuration.



MN flag

Meaning: **M**ono/**N**eutro

Presence of a significantly large population of cells located in the separation threshold area between monocytes and neutrophils. This flag occurs when the number of particles counted in this area is higher than the limit set up in **MN#** or the number of particles counted in MN versus the total number of WBC is above the **MN%** limit.

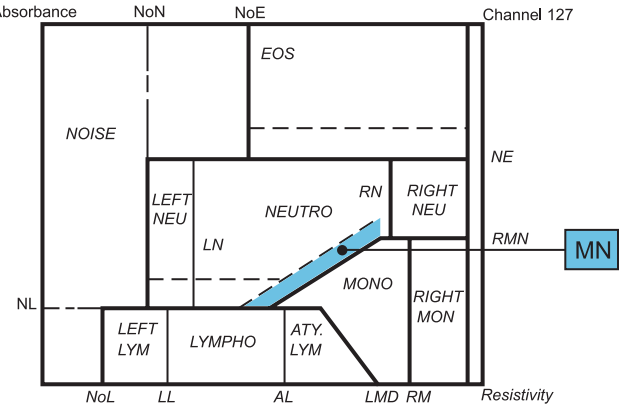
Suspected abnormalities:
- Monocytes having granules in their cytoplasm or hyperbasophilic monocytes,
- Young neutrophils with non-segmented nuclei (bandcells).

This flag occurs associated with an (**!**) on:
- ALY % ALY #
- LIC % LIC #

and replaces:
- NEU %, NEU #, MON %, MON # by ----.

+ ***Standard** values for **MN**:*
% 100
 # 120

Adjustment:
see Chapter 4. Instrument configuration.



LN flag

Meaning: **L**eft **N**eutro

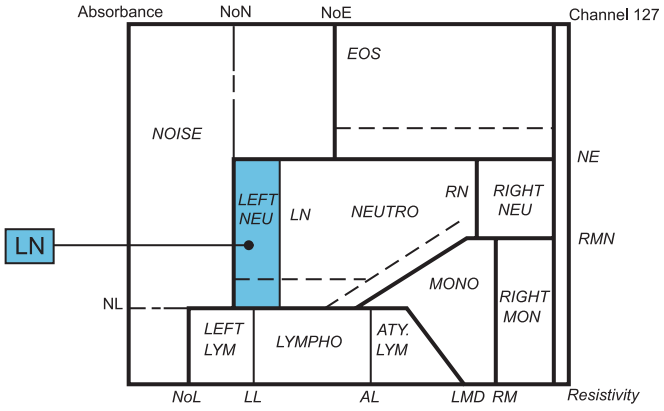
Presence of a significantly large population of cells located on the left-hand side of the neutrophil area. This flag occurs when the number of particles counted in this area is higher than the limit setup in **LN#** or when the number of particles counted regarding the total number of WBC is above **LN%** limit.

Suspected abnormalities:
- Neutrophil destruction due to incorrect storage of the sample or an old sample,
- Contamination, stroma or platelet aggregates.

This flag occurs associated with an (**!**) on all WBC differencial parameters.

+ ***Standard** values for **LN**:*
% 2.5
 # 999

Adjustment:
see Chapter 4. Instrument configuration.



NE flag

Meaning: **N**eutro/**E**osino.

Presence of a significantly large population of cells located in the separation area between neutrophils and eosinophils because of a superimposition of the 2 populations. This flag occurs when the number of particles counted in this area is higher than the limit setup in **NE#** or when the number of particules counted regarding the total number of WBC is above the **NE%** limit.

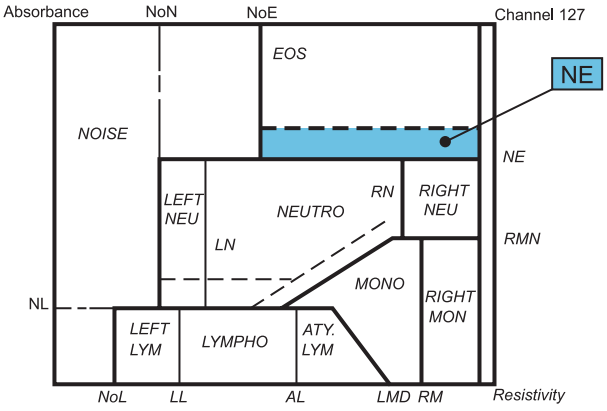
Suspected abnormalities:
- Young eosinophils,
- Giant hypersegmented neutrophils,
- Eosinophils with low intracytoplasmic material,
- Immature cells.

This flag is associated with an (**!**) on:
- LIC % LIC #

and replaces:
- NEU %, NEU #, EOS %, EOS # by ----.

+ ***Standard** values for **NE**:*
% 1.1
 # 60

Adjustment:
see Chapter 4. Instrument configuration.



ALY flag

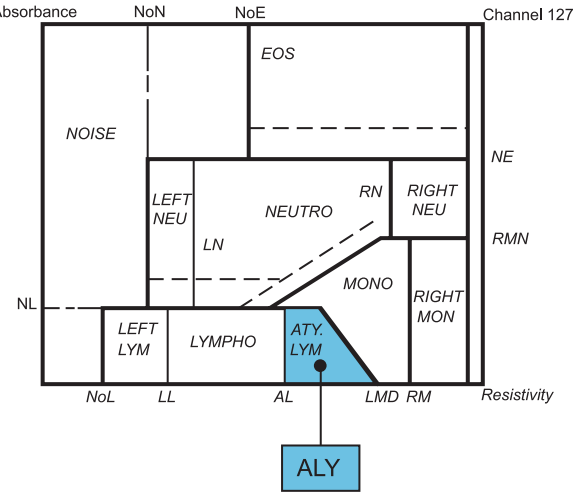
Meaning: **A**typical **L**ymphocytes

Presence of a significantly large population of cells located on the right-hand side of the Lymphocytes area. This flag occurs when the number of particles counted in this area is higher than the limit setup in **ALY#** or when the number of particles counted regarding the total number of WBC is above the **ALY%** limit.

Suspected abnormalities:
- Large Lymphocytes,
- Reactive Lymphoid forms,
- Stimulated Lymphocytes,
- Plasmocytes.

+ ***Standard** values for **ALY**:*
% 2
 # 0.2

Adjustment:
see Chapter 4. Instrument configuration.



RM flag

Meaning: **R**ight **M**ono

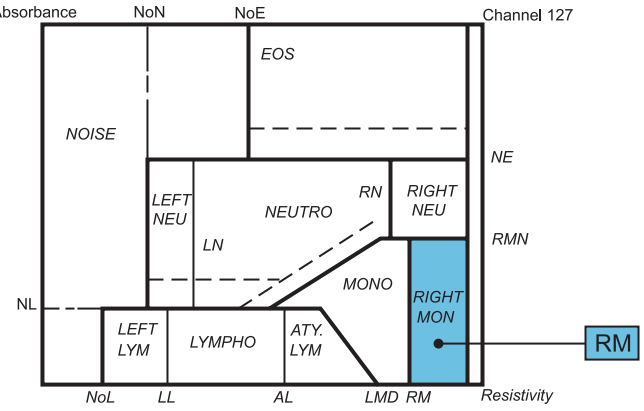
Presence of a significantly large population of cells located on the right-hand side of the monocyte area (low LIC). This flag occurs when the number of particles counted in this area is higher than the limit setup in **RM#** or when the counted particles regarding the total of WBC is above **RM%** limit.

Suspected abnormalities:
- Large monocytes,
- Hyperbasophilic monocytes,
- Myelocytes or promyelocytes,
- Large blasts.

This flag occurs associated with an (**!**) on:
- NEU % NEU #
- MON % MON #
- LIC % LIC #

+ ***Standard** values for **RM**:*
% 1.1
 # 999

Adjustment:
see Chapter 4. Instrument configuration.



RN flag

Meaning: **R**ight **N**eutro

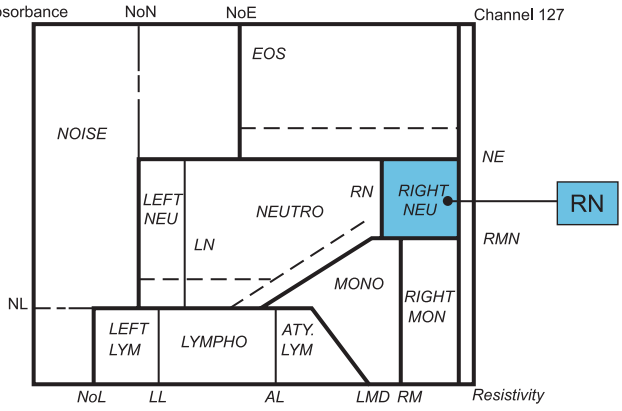
Presence of a significantly large population of cells located on the right-hand side of the neutrophil area (high LIC). This flag occurs when the number of particles counted in this area is higher than the limit setup in **RN#** or when the number of particles counted regarding the total number of WBC is above the **RN%** limit.

Suspected abnormalities:
- Large neutrophils,
- Immature cells from granulocyte hemopoiesis (metamyelocytes, myelocytes, promyelocytes).

This flag is associated with an (**!**) on:
- NEU % NEU #
- LIC % LIC #

+ ***Standard** values for **RN**:*
% 1.1
 # 999

Adjustment:
see Chapter 4. Instrument configuration.



LIC flag

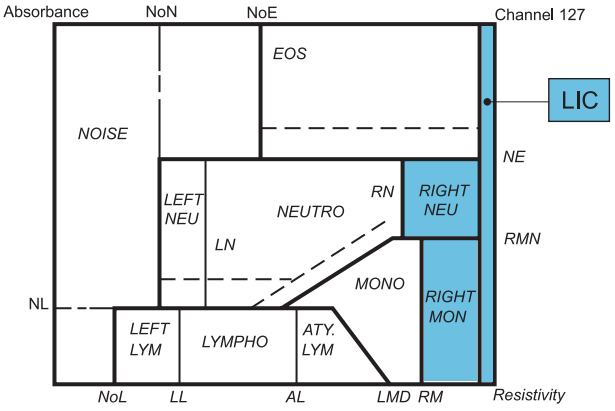
Meaning: **L**arge **I**mmature **C**ells

Presence of a significantly large population of cells located on **RN** + **RM** + **channel 127** areas. This flag occurs when the number of particles counted in this area is higher than the limit set up in **LIC#**, or when the number of counted particles regarding to the total number of WBC is above the **LIC%** limit.

Suspected abnormalities:
- Large monocytes,
- Hyper basophilic monocytes,
- Myelocytes, Metamyelocytes, Promyelocytes,
- Large blasts,
- Large neutrophils.

+ ***Standard** values for **LIC**:*
% 2
 # 0.2

Adjustment:
see Chapter 4. Instrument configuration.



5.3.3. Flags on WBC/BASO histogram

CBC and DIFF Mode

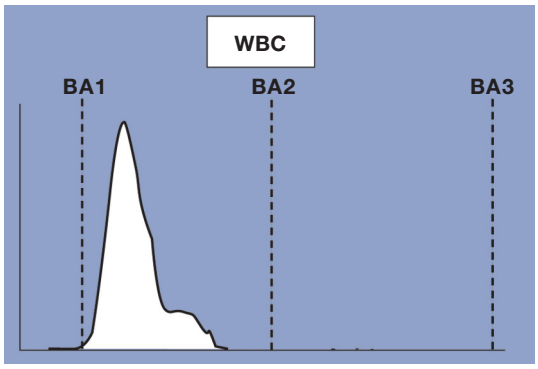
L1 flag is established according to the ratio of the cells counted between the 0 channel and BA1.

L1 indicates the presence of an abnormal number of cells in comparison with leukocytes.

Suspected abnormalities:
- Plt aggregates,
- NRBCs.

+ ***Standard** values for **L1**:*
% 3
 # 200

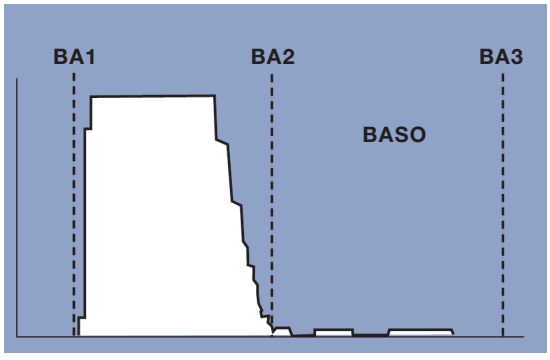
Adjustment:
see Chapter 4. Instrument configuration.



DIFF mode only

MB (Meaning: **M**ono **B**aso)
This flag occurs when the percentage of basophils found in the Baso channel is above the percentage of Lympho/Mono/Neutro raw counts found on the matrix channel.

BASO+
If the BASO % exceeds 50%, a **BASO+** flag is generated.
The Basophils are not taken away from the matrix populations and ---- is displayed instead of the BAS % and BAS #.



5.3.4. Flags on RBC histogram

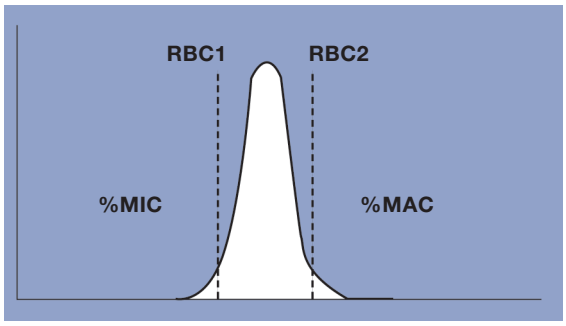
MIC & MAC flags

MIC and **MAC** flags are generated when the percentage of cells counted in the microcytic area (MIC) and macrocytic area (MAC) compared to the total number of RBCs are above the limits set up by user.

• RBC1 and RBC2 thresholds define the microcytic and macrocytic areas and are calculated according to the MCV and the RDW.

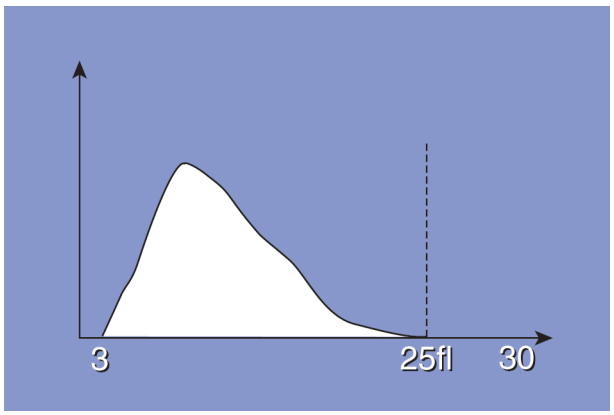
+ ***Standard** values for*
***Mic**: % 5*
***Mac**: % 45*

Adjustment:
see Chapter 4. Instrument configuration.



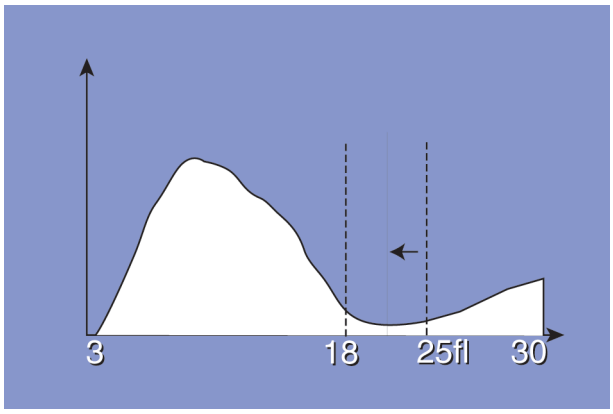
5.3.5. Flags on Plt histogram

• The Plt histogram has 256 channels between 2fL and 30fL. A mobile threshold (at 25fL by default) moves according to the microcytic RBCs present in the platelet analysis area.

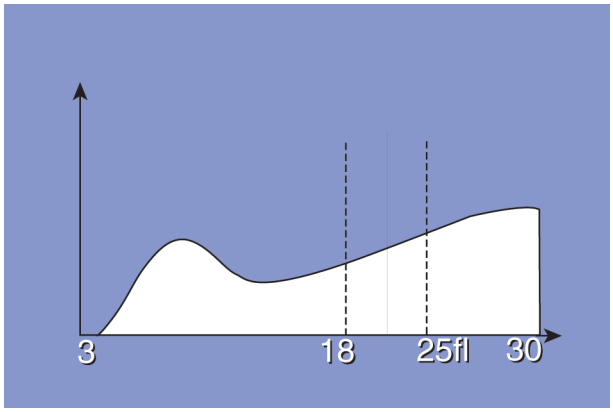


• The Plt flags are generated under the following conditions:

+ ***An*** *excessive presence of particles on the right side of the threshold area (after 25fL) will generate the* ***MIC*** *(****Mic****rocytes) flag (shown in the platelet alarm area). In this case the mobile threshold looks for a valley between 18fL and 25fL (standard area).*

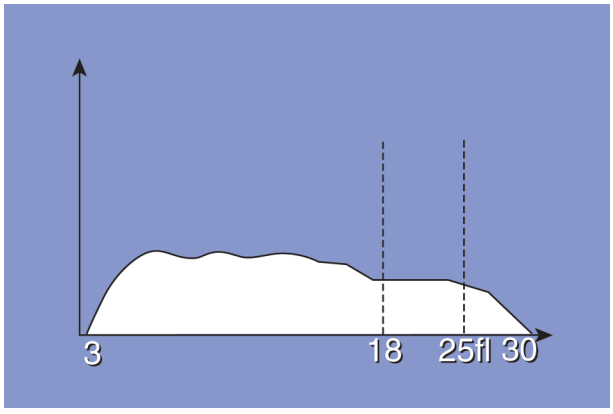


+ ***When*** *the mobile threshold can not be positioned in the standard area (from 18fL to 25fL), a* ***Plt*** *reject (*) and a* ***MIC*** *flag are generated. The Plt result is not reliable. Verify the result using a Platelet Rich Plasma (PRP) or a manual count.*



+ ***If*** *the mobile threshold cannot be positioned (no valley between the Plt and RBC histograms) the* ***SCH*** *(****Sch****izocytes) flag is generated.*

Suspected abnormalities:
• *Presence of schizocytes,*
• *Presence of Platelet aggregates, check the result on a slide.*



+ **SCL** (**S**mall **C**ell) indicates the presence of small cells in the 2fL and 3fL zone. A second analysis should be carried out and the results verified.

5.3.6. WBC balance

+ ***WBC Balance*** *can be activated or deactivated: see Chapter 4. Instrument configuration.*

Control of the injected volume in the optical flowcell allows a second count of the WBC.

The two counts are compared, if the difference between both counts is higher than a defined threshold (depending of the quantity measured), a **LMNE+** or a **LMNE-** flag is generated.

- WBC count is within 0 and 2501:
If the WBC LMNE count is higher than 50% of the WBC BASO count, a **LMNE+** flag is generated.
If the WBC LMNE count is lower than 50% of the WBC BASO count, a **LMNE-** flag is generated.

- WBC count is within 2501 and 8000:
If the WBC LMNE count is higher than 20% of the WBC BASO count, a **LMNE+** flag is generated.
If the WBC LMNE count is lower than 20% of the WBC BASO count, a **LMNE-** flag is generated.

- WBC count is higher than 8000:
If the WBC LMNE count is higher than 15% of the WBC BASO count, a **LMNE+** flag is generated.
If the WBC LMNE count is lower than 15% of the WBC BASO count, a **LMNE-** flag is generated.

The WBC BASO channel is considered as a reference and is used to calibrate the WBC LMNE channel. The ratio calculated between the two channel calibration coefficients is, except technical intervention, stable. In any case it is the WBC BASO result that is reported.

NOTE

The WBC balance flags (LMNE+ and LMNE-) shall not be triggered if and only if:
- ***The test selected is «CBC».***
- ***The WBC Balance option is not activated.***

These flags are associated with an (!) on all differential parameters (% and #).
L1 flag is associated with an (!) on WBC value and on absolute values of the differential parameters.

5.4. Pathology messages

• Pathology suspicion messages may be displayed and printed out. Triggering conditions are linked to the laboratory limits entered by the user.

WARNING

These messages indicate a possible pathological disorder and should be used to assist with quick and efficient screening of abnormal samples and for diagnosis. It is recommended to use suitable reference methods to confirm diagnoses.

NOTE

There is no pathological message in the following cases:

- ***On WBC: For a WBC < 0.1x10³mm³ or WBC > 91.3x10³/mm³ or for a counting reject (except if an Erythroblast flag is the NUCLEATED RBC).***
- ***On RBC: For a counting reject or an RBC value < 0.1x10⁶/mm³.***
- ***On Plt: For a counting reject or a Plt value < 5x10³/mm³.***

5.4.1. WBC messages

«H»: high extreme limit

«L»: low extreme limit

Message	Triggering Condition
LEUKOCYTOSIS	WBC > WBC H
LEUKOPENIA	WBC < WBC L
LYMPHOCYTOSIS	* LYM # > LYM # H or if LYM % > LYM % H
LYMPHOPENIA	* LYM # < LYM # L or if LYM % < LYM % L
NEUTROPHILIA	* NEU # > NEU # H or if NEU % > NEU % H
NEUTROPENIA	* NEU # < NEU # L or if NEU % < NEU % L
EOSINOPHILIA	* EOS # > EOS # H or if EOS % > EOS % H
MYELEMIA	NEU % > NEU % H and LIC # > LIC # H
LARGE IMMATURE CELL	LIC # > LIC # H or LIC % > LIC % H
ATYPIC LYMPHOCYTE	ALY # > ALY # H or ALY % > ALY % H
LEFT SHIFT	(MN or NL) and RN
NUCLEATED RBC	LL
MONOCYTOSIS	* MON # > MON # H or if MON % > MON % H
BASOPHILIA	* BAS # > BAS # H or if BAS % > BAS % H
BLASTS	BAS # > BAS # H and LIC # > LIC # H and RM

** means the pathology is detected, firstly, on the high and low absolute values of the corresponding parameter.*

5.4.2. RBC messages

«H»: high extreme limit

«L»: low extreme limit

Message	Triggering Conditions
ANEMIA	Hgb < Hgb L
ANISOCYTOSIS	RDW > RDW H
MICROCYTE	on MIC flag
MICROCYTE+	% MIC > 10 %
MICROCYTE++	% MIC > 15 %
MACROCYTE	on MAC flag
HYPOCHROMIA	MCHC < MCHC L
COLD AGGLUTININ	MCHC > MCHC H and WBC < 91.3x10³/mm³
MICROCYTOSIS	MCV < MCV L
MACROCYTOSIS	MCV > MCV H
ERYTROCYTOSIS	RBC > RBC H

5.4.3. Plt messages

«H»: high extreme limit

«L»: low extreme limit

Message	Triggering Conditions
THROMBOCYTOSIS	Plt > Plt H
THROMBOCYTOPENIA	Plt < Plt L
MICROCYTOSIS SCHIZOCYTE SMALL CELL	See Triggering conditions for these flags in paragraph ***Flags on RBC curve***
Plt AGGREGATE (1)	Plt < 150x10³/mm³ + WBC Reject or NO + PDW > 20 or NO + MPV > 10 or NO + Plt < 150x10³/mm³ or NO + WBC Reject or L1 or LL1 + PDW > 20 or L1 or LL1 + MPV > 10 or L1 or LL1 + Plt < 250x10³/mm³
ERYTHROBLASTS (2)	LL or WBC reject + L1 or WBC reject + LL1
ERYTHROBLASTS Plt AGGREGATE	(1) and (2) are false + (L1 or LL1 or WBC reject)
MACROPLATELETS	MPV > 11

5.4.4. Miscellaneous

«H»: high extreme limit

«L»: low extreme limit

MESSAGE	TRIGGERING CONDITIONS
PANCYTOPENIA	WBC < WBC L and RBC < RBC L and Plt < Plt L

5.5. Analyzer alarms

Total rejection of the matrix

Meaning: poor correlation

The percentage of validated cells is abnormally low, appears when the correlation between the resistivity measurement of particles and their optical measurement is less than 50%.

Suspected abnormalities:
- Stroma interfering with measurement,
- Strong pollution,
- Incorrect adjustment of the optical bench.

Others

- From LMNE Matrix: **NO** flag
- From WBC Balance: **LMNE+**; **LMNE-**
- From WBC/BASO Histogram: **BASO+**

6. CALIBRATION

- Two modes are available:

1. AUTOCALIBRATION: using calibration blood samples.
2. CHANGE COEFFICIENT: calibration coefficients are known and can be entered directly.

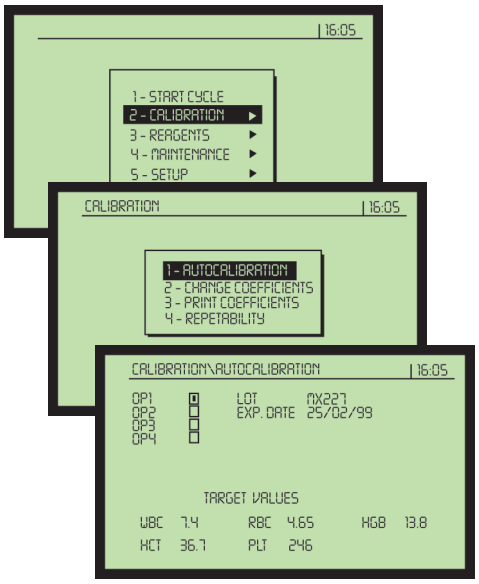
6.1. Autocalibration

- When the «***3. Calibration verification***» has failed, calibrate the instrument as follows:

6.1.1. Operator selection

- Enter the «*AUTOCALIBRATION*» menu.
- Move the cursor to one of the 4 required operator identifications.
- Press . to validate the selection.
- See *Chapter 4. Instrument configuration* to change the operator identification.

6.1.2. Change lot number



- Move the cursor to «LOT ....» field if this one has to be changed. If not perform the «***6.1.3. Change expiration date***».
- Press Enter: the cursor turns to a flashing «_» below the figures or letters to replace.
- Use ↓ and ↑ to display the letters and numerical keyboard to display the figures.
- Press Enter to validate the lot number.

6.1.3. Change expiration date

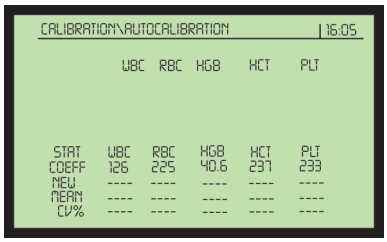
- Move to the «EXP DATE» field if this one has to be changed. If not perform the «***6.1.4. Change target values***».
- Proceed as described in «***6.1.2. Change lot number***» to enter the new date.
- See *Chapter 4. Instrument configuration* to change the date format.

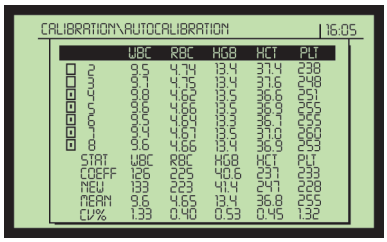
6.1.4. Change target values

- Move to the «TARGET» field if the this one has to be changed. If not press ESC directly.
- Enter the new WBC target value if required, and press Enter.
- The menu turns to RBC target value.
- Repeat the same procedure for RBC, HGB, HCT, PLT.
- When this last value has been modified press ESC: the calibration results chart table is displayed.

6.1.5. Run calibration

- Prepare the calibrator according to the specific instructions (temperature, mixing, etc...).
- Open the vial and position the sampling needle deeply inside the bottle.
- Press the sampling bar located behind the needle.
- When the cycle LED stops flashing, remove the vial and replace the cap on the calibrator.
- When the analysis cycle ends, the first result is displayed on the result chart table.
- Run the second calibrator sample.

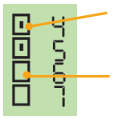




- The calibration of the *ABX PENTRA 60* can be performed on 3 to 11 analyses.
- The autocalculation module performs statistics on these results in order to obtain the best calibration coefficients.

IMPORTANT

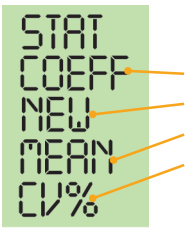
Risk of erroneous results if the calibrator is not continuously mixed between each analysis. Continue mixing the calibrator between each analysis.
In order to obtain the best possible calibration, it is recommended to run at least 5 calibrator samplings.



Interpretation:

• Currently, each result is selected: i.e. that this one is involved in the statiscal calculation.

• To discard a result from the statiscal calculation, move the cursor to the result line and press . to obtain a single square.



• Use the ↓ and ↑ to scroll up and down on the results chart table.

• The instrument calculates the statistical calibration factors for each parameter:

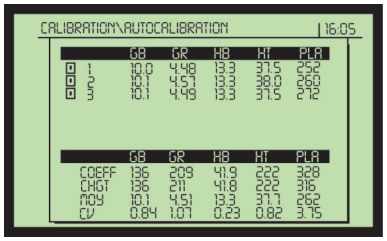
- Recall of the previous coefficient of calibration
- Calculation of the new coefficient of calibration
- Mean of the results
- Coefficient of variation

• Press ESC.

A- Calibration passed

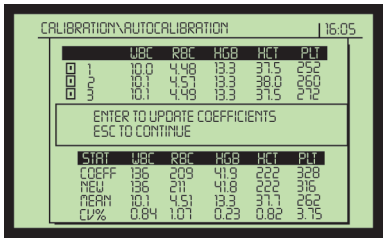
• If the statistic figures are within the acceptable limits:

- Coefficient of variation is within the limits setup as described *Chapter 4. Instrument configuration* and,
- The percentage difference between the target and the mean value is less than 20.

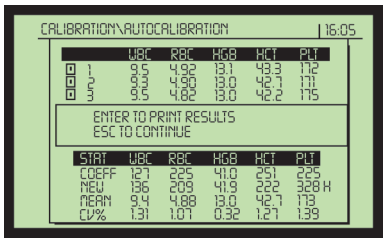


The calibration passed:

• Press ↵ Enter to save the new coefficients.



• Press ↵ Enter to print the new coefficients.



The results involved in the statistical calculations are shown wih a «X» in the first column.

CALIBRATION

25/02/99 18:05:39

CALIBRATION DATE 25/02/99 OPERATOR OP1

LOT# JX94N EXP. DATE 05/05/99

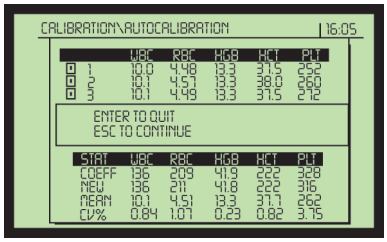
TARGET VALUES

WBC	10.1	$10^3/mm^3$
RBC	4.54	$10^6/mm^3$
HGB	13.3	g/dl
HCT	37.7	%
PLT	252	$10^3/mm^3$

		WBC	RBC	HGB	HCT	PLT
X	1	10.0	4.48	13.3	37.5	252
X	2	10.1	4.57	13.3	38.0	260
X	3	10.1	4.49	13.3	37.5	272

	WBC	RBC	HGB	HCT	PLT
PREV. COEFF	136	209	41.9	222	328
COEFF	136	211	41.8	222	316
MEAN	10.1	4.51	13.3	37.7	262
CV	0.84	1.07	0.23	0.82	3.75

• The calibration is completed, press Enter to quit the calibration chart table.

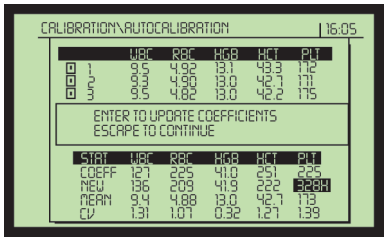


B- Calibration forced

If the statistic figures are not within the acceptable limits:

• Coefficient of variation is not within the limits or,

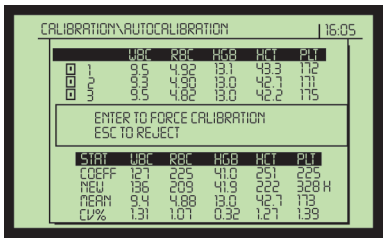
• The percentage difference between the target and the mean value is greater than 20.



• The out of range coefficient is shown in reverse video and a flag «H» (higher) or «L» (lower) is displayed next to it.

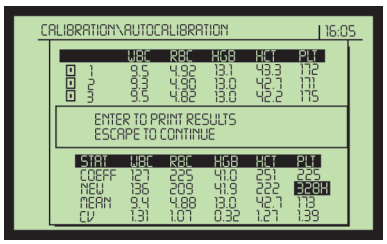
• Press Enter to save the new coefficients: **the calibration is forced**,

(or ESC to quit and save the previous coefficients).


CALIBRATION\AUTOCALIBRATION
16:05
WBC RBC HGB HCT PLT
1 9.5 4.92 13.1 43.3 172
2 9.3 4.90 13.0 42.7 171
3 9.5 4.82 13.0 42.2 175
ENTER TO FORCE CALIBRATION
ESC TO REJECT
STAT WBC RBC HGB HCT PLT
COEFF 127 225 41.0 251 225
NEW 136 209 41.9 222 328 H
MEAN 9.4 4.88 13.0 42.7 173
CV% 1.31 1.07 0.32 1.27 1.39

• Press Enter to print the new coefficients: a message «**Forced calibration**» is printed out on the calibration print form.

• Press Enter to quit the calibration chart table.


CALIBRATION\AUTOCALIBRATION
16:05
WBC RBC HGB HCT PLT
1 9.5 4.92 13.1 43.3 172
2 9.3 4.90 13.0 42.7 171
3 9.5 4.82 13.0 42.2 175
ENTER TO PRINT RESULTS
ESCAPE TO CONTINUE
STAT WBC RBC HGB HCT PLT
COEFF 127 225 41.0 251 225
NEW 136 209 41.9 222 328H
MEAN 9.4 4.88 13.0 42.7 173
CV 1.31 1.07 0.32 1.27 1.39

The results involved in the statistical calculations are shown wih a «*X*» in the first column.

CALIBRATION

25/02/99 17:55:24

CALIBRATION DATE 25/02/99 OPERATOR OP1

FORCED CALIBRATION

LOT # JX94N EXP. DATE 05/05/99

TARGET VALUES

WBC	10.1	$10^3/mm^3$
RBC	4.54	$10^6/mm^3$
HGB	13.3	g/dl
HCT	37.7	%
PLT	252	$10^3/mm^3$

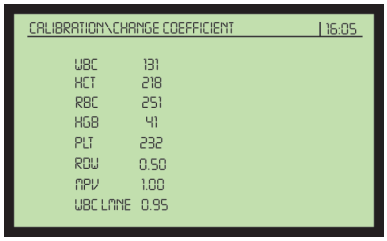
		WBC	RBC	HGB	HCT	PLT
X	1	9.5	4.92	13.1	43.3	172
X	2	9.3	4.90	13.0	42.7	171
X	3	9.5	4.82	13.0	42.2	175

	WBC	RBC	HGB	HCT	PLT
PREV. COEFF	127	225	41.0	251	225
COEFF	136	209	41.9	222	328 H
MEAN	9.4	4.88	13.0	42.7	173
CV	1.31	1.07	0.32	1.27	1.39

6.2. Change coefficients

• Calibration can be achieved directly by changing the calibration coefficients when they are known.

• Move the cursor to function «CHANGE COEFFICIENTS» and press Enter.



• A specific password is requested to enter the function.
• Enter the «User password» (defined as described *Chapter 4. Instrument configuration*) and press Enter.
• Move the cursor down to the WBC, RBC, HGB, HCT, PLT and RDW positions and enter the required new coefficients.

NOTE

As the WBC LMNE parameter is automatically calibrated by the WBC balance (see section 4 - WBC balance), only an HORIBA ABX certified technician is allowed to change it.

• When the required coefficients have been changed, press Enter to record the setup.

CALIBRATION COEFFICIENT	STD VALUE	MINIMUM	MAXIMUM
WBC	137	90	200
RBC	225	160	290
HGB	40.0	25.0	55.0
HCT	220	160	290
PLT	290	180	400
RDW	0.35	0.1	0.9
MPV	1.00	0.1	10

• RDW can be calibrated by means of calibration coefficients. These coefficients are incremented to 0.35 by default. The RDW is calculated according to the below formula:

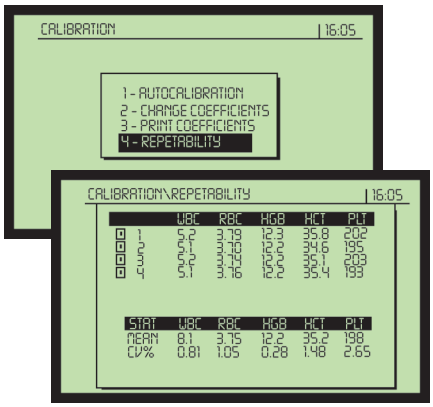
RDW result = RDW coeff x RDW calculated

6.3. Print coefficients

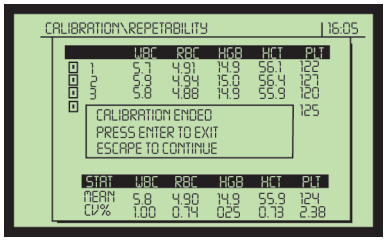
• Move to «PRINT COEFF» and press Enter to print out the coefficients.

6.4. Repeatability

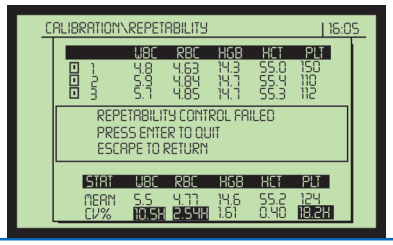
• The repeatability check is done by doing fresh blood CBC runs (from 3 to 11 maximum). Means and CV, for each parameter, are displayed into menu CALIBRATION\REPEATABILITY.
• Open «REPEATABILITY» menu.
• To run specimens and to select/unselect results (used for the statistic calculation), proceed as described into «***6.1.5. Autocalibration***».


CALIBRATION
16:05
1 - AUTOCALIBRATION
2 - CHANGE COEFFICIENTS
3 - PRINT COEFFICIENTS
4 - REPETABILITY
CALIBRATION\REPETABILITY
16:05
WBC RBC HGB HCT PLT
1 5.2 3.19 12.3 35.8 202
2 5.1 3.10 12.2 34.6 195
3 5.2 3.14 12.2 35.1 203
4 5.1 3.16 12.2 35.4 193
STAT WBC RBC HGB HCT PLT
MEAN 8.1 3.15 12.2 35.2 198
CV% 0.81 1.05 0.28 1.48 2.65

• Press Enter when analysis are completed.
• If CV results are lower than the defined limits, a message is displayed (as shown on the screen). When leaving the «REPEATABILITY» menu, results and statistics are erased.


CALIBRATION\REPETABILITY
16:05
WBC RBC HGB HCT PLT
1 5.7 4.91 14.9 56.1 122
2 5.8 4.94 15.0 56.4 127
3 5.8 4.88 14.9 55.9 120
125
CALIBRATION ENDED
PRESS ENTER TO EXIT
ESCAPE TO CONTINUE
STAT WBC RBC HGB HCT PLT
MEAN 5.8 4.90 14.9 55.9 124
CV% 1.00 0.74 0.25 0.73 2.38

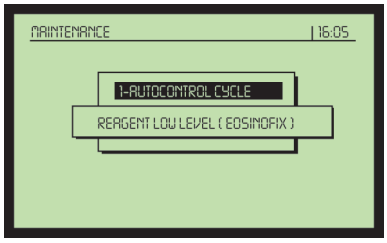
• If CV results are out of range, they are displayed in reverse video and a flag H is displayed next to it. CV limits are the one used for the Calibration and can be changed as described *Chapter 4. Instrument configuration* ***Calibration***.


CALIBRATION\REPETABILITY
16:05
WBC RBC HGB HCT PLT
1 4.8 4.63 14.3 55.0 150
2 5.9 4.84 14.7 55.4 110
3 5.7 4.85 14.7 55.3 112
REPETABILITY CONTROL FAILED
PRESS ENTER TO QUIT
ESCAPE TO RETURN
STAT WBC RBC HGB HCT PLT
MEAN 5.5 4.77 14.6 55.2 124
CV% 10.5H 2.54H 1.61 0.40 18.2H

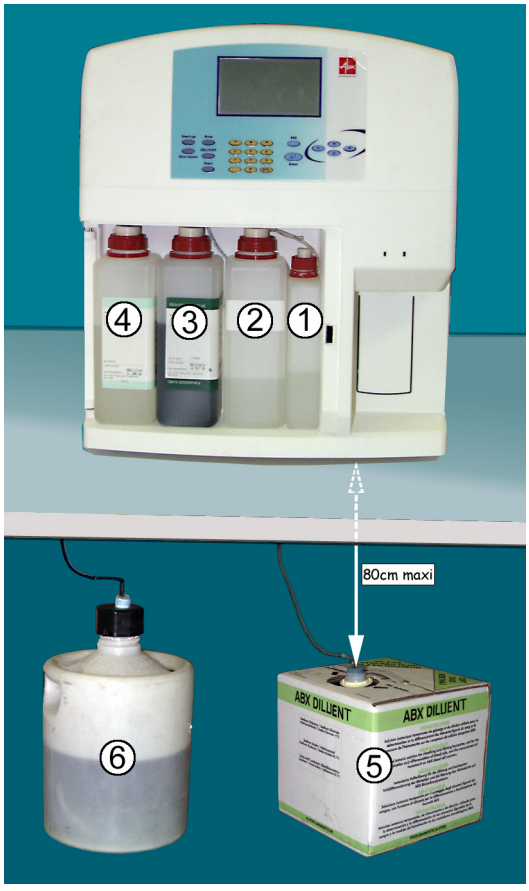
7. REAGENTS LEVEL/CHANGE

• The instrument calculates each reagent bottle capacity according to the number of cycles run. The instrument performs a reagent capacity check on each cycle. If a reagent low level is expected, an alarm is triggered off.

Example for the *ABX* ***EOSINOFIX***: «REAGENT LOW LEVEL (EOSINOFIX)».



7.1. Reagents connections



1 - *ABX* ***LYSE***
2 - *ABX* ***BASOLYSE II***
3 - *ABX* ***EOSINOFIX***
4 - *ABX* ***CLEANER***
5 - *ABX* ***DILUENT***
6 - WASTE CONTAINER

CAUTION

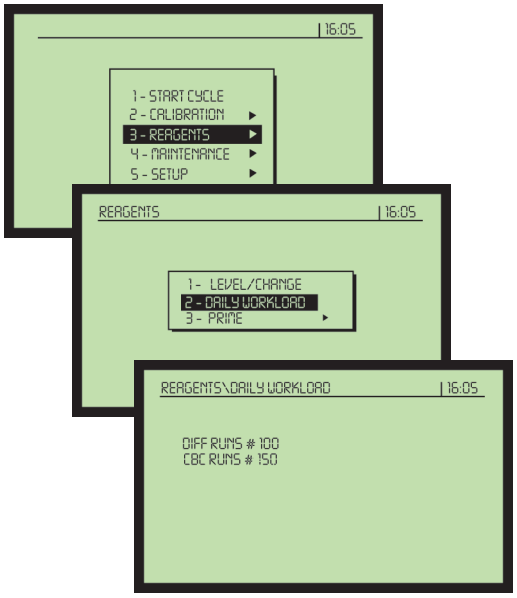
The container must not be installed further than 80cm from the instrument. The diluent input tubing is cristal 3 x 6 (2 meters maximum), and the waste output tubing is cristal 4 x 6 (2 meters maximum).

7.2. Daily workload

• The operator can entered the approximated number of CBC and DIFF analyses performed each day.

• This «daily workload» warns the operator if a reagent low level is expected during the day. In this case, an alarm is triggered out: «REAGENT LOW LEVEL» at the end of the Startup cycle.

• The operator can replace the bottle (or container) immediately or performed the analyses until the specific reagent alarm is triggered (see ***7.3. Reagent replacement***).

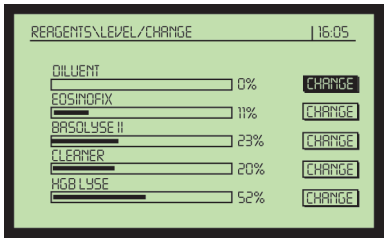


• Ranges are given in the below chart table:

Mode	Default Value	Min.	Max.
DIF	40	1	500
CBC	10	1	500

7.3. Reagent replacement

- Open the reagent «LEVEL/CHANGE» menu.
- If a reagent level indicates «0%», this one has to be replaced.



IMPORTANT

Risk of erroneous results if one reagent is poured to another container. Never pour reagents from one container to another. Particles at the bottom of the old container can contaminate the new reagent and will cause unacceptable background results especially for Plts.

Properly dispose of the empty reagent bottle.

- Replace the bottle (or diluent container) as follows.

7.3.1. Bottle replacement

- Open the reagent front door.
- Remove the empty bottle from the reagent compartment.
- Unscrew the «bottle stopper and reagent straw» assembly.
- Replace the bottle by a new one and screw the «stopper/straw» back on the new bottle.

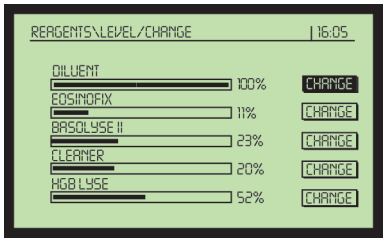
- Proceed as described in «***7.3.3. Level update***».

7.3.2. Diluent container replacement

- Remove the diluent straw.
- Replace the container by a new one and install the straw deep in the new container.
- Proceed as described in «***7.3.3. Level update***».

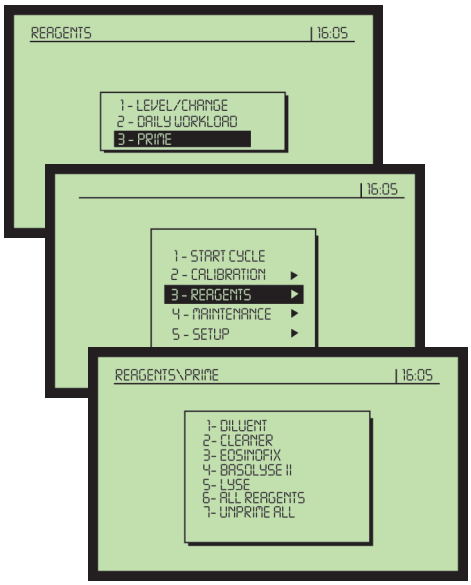
7.3.3. Level update

- Use the ↓ and ↑ to display the reagent «CHANGE» key in reverse video.
- Press Enter: a prime cycle is automatically run.
- The reagent level is updated to «100%».



7.4. Prime

- This function runs priming cycles useful at the instrument first installation or in case of a technician intervention.



CAUTION

Remove the straws from the reagent bottles and containers before running a «7- Unprime all» cycle.

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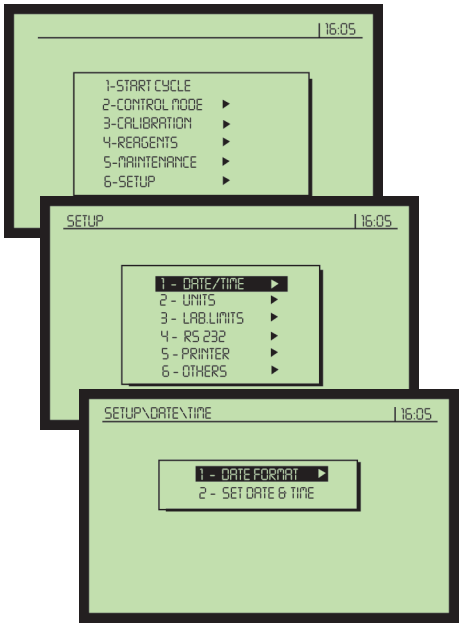
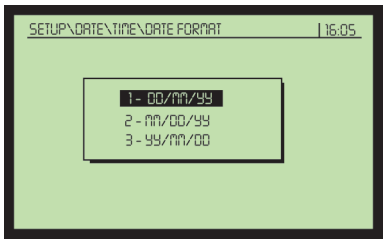
1. DATE AND TIME

- Date and time can be setup according to the country specifications:

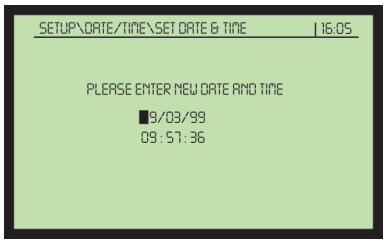


1.1. Date format



- 3 different date formats can be used:
DD.MM.YY
MM.YY.DD
YY.MM.DD
- Move the cursor in front of the required selection and press Enter.
- The new date format is recorded.

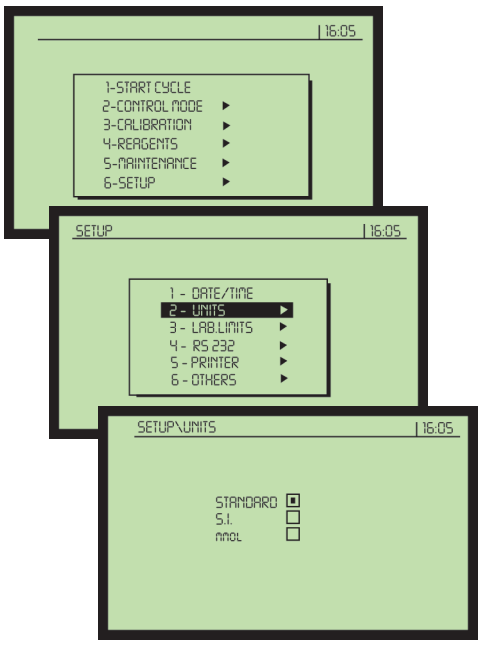
1.2. Change date



- Open the «*SET DATE & TIME*» menu.
- Type in the new date and (or) time.
- Press Enter to confirm.

2. UNITS

• The operator has the choice between 3 different unit systems, move the cursor in front of the required unit system and press .



• Press ESC to exit.

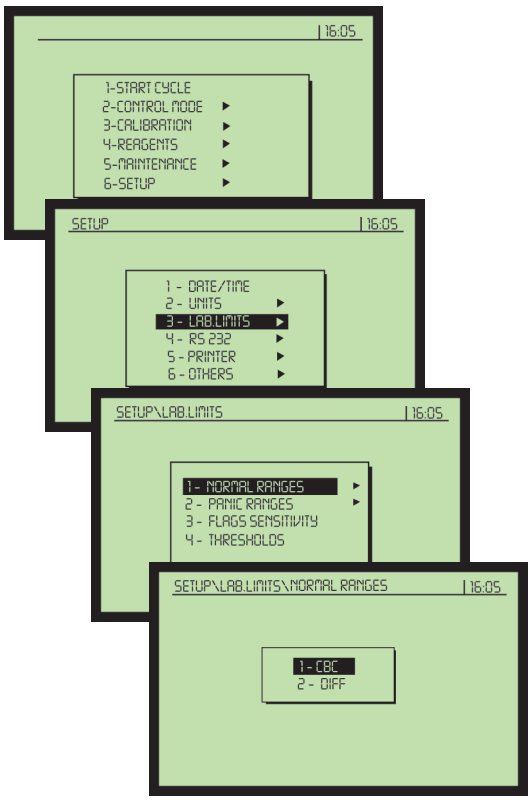
3. LABORATORY LIMITS

• Laboratory limits can be set by the operator according to its own specifications.

3.1. Normal ranges

• Results that exceed the «***Normal ranges***» limits are identified with a flag:
«**h**» for results above the upper limit,
«**l**» for results below the lower limit.

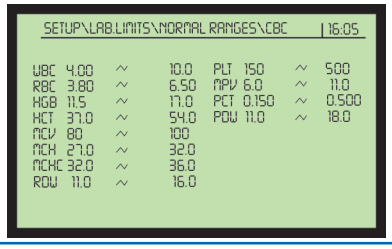
• **CBC's** and **differential normal ranges** are separated between the two following screens.



3.1.1. CBC's Normal ranges

• The CBC's normal ranges are factory adjusted to the following values. To modify them move the cursor to one of them and enter the new value.

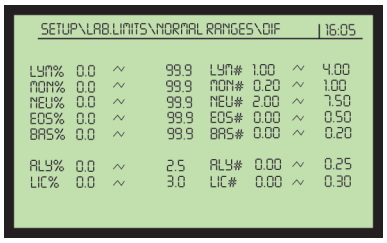
• Step to the next one, etc... Press Enter to validate.



3.1.2. DIF's Normal ranges

• The DIFF normal ranges are factory adjusted to the following values. To modify them move the cursor to one of them and enter the new value.

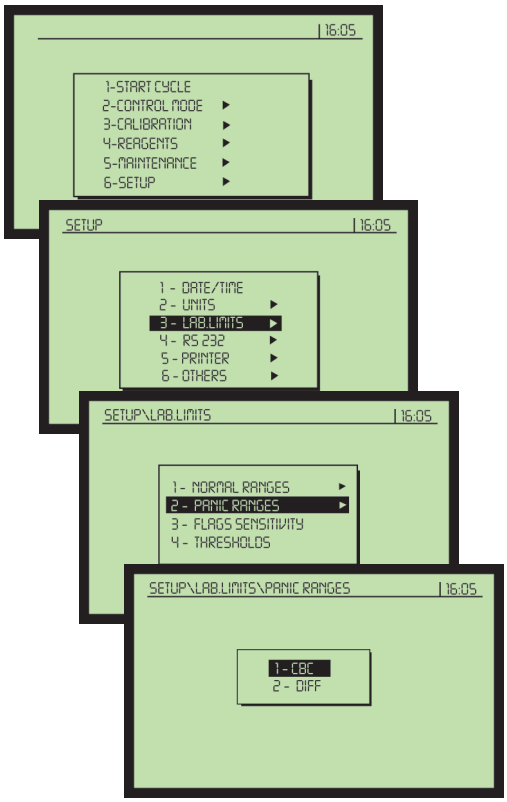
• Step to the next one, etc... Press Enter to validate.



3.2. Panic ranges

• Results that exceed the «***Panic ranges***» limits are identified with a flag:
«**H**» for results above the upper limit,
«**L**» for results below the lower limit.

• **CBC's** and **differential panic ranges** are separated between two screens. See «***1.2.1. Normal ranges***» to adjust them.

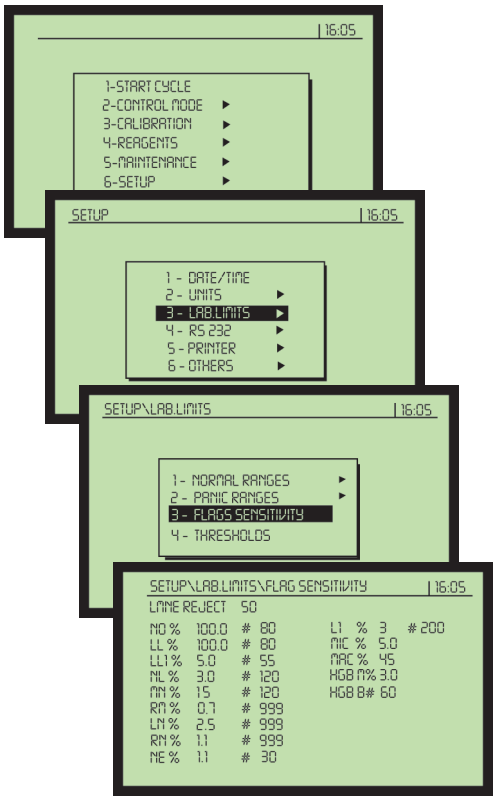


3.3. Flag Sensitivity

• Each flag is adjustable according to a percentage and (or) an absolute value. Beyond these values the corresponding flag is triggered off.

• The standard values are factory installed on the instrument and shown in the screen below.
Move the cursor to the value to modify and enter the new value (Password is required to enter this function).

• Press Enter to validate.



• The following chart table gives the standards:

FLAG SENSITIVITY STANDARDS		
FLAGS	%	#
No	100	120
LI	100	50
LI1	5	45
NI	3	120
Mn	100	120
Rm	1.1	999
Ln	2.5	999
Rn	1.1	999
Ne	1.1	60
L1	3	200
Mic	5	—
Mac	45	—
HGB M	3.0	—
HGB B	—	60

NOTE

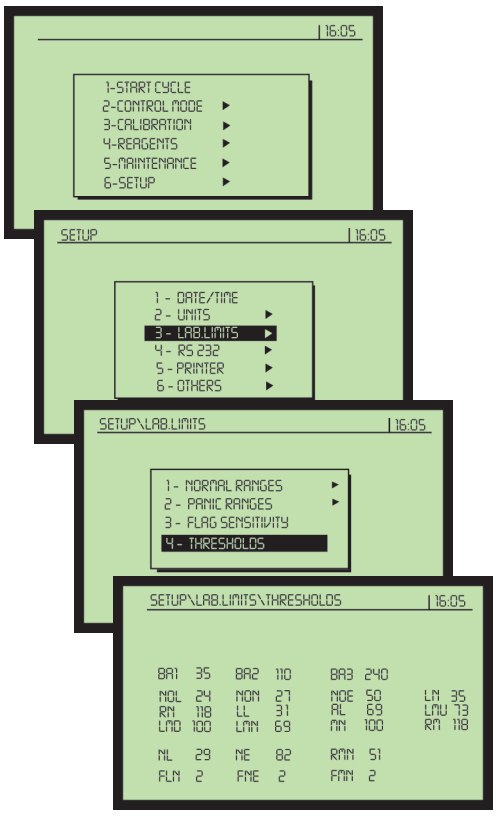
The meaning of each flag is described Chapter 3. Specimen Run & Results.
MHGB% indicates the reject level between the three HGB measures.
BHGB# allows to reject an HGB reference value that is faulty.
(see Chapter 3. Specimen Run & Results)

3.4. Thresholds

3.4.1. WBC/BASO

• All of the leukocytes are shown between the BA1 and BA3 thresholds.
L1 absolute value is calculated between the channel 0 and the BA1 threshold (see *Chapter 3. Specimen Run & Results*).

• The percentage of basophils is calculated according to the number of particles from the BA2 threshold to the BA3 threshold.

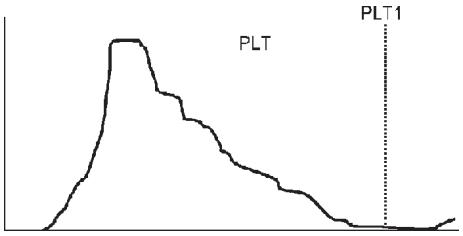


• These thresholds are factory adjusted to the following values:

Threshold	Purpose	Channel
0	L1# counting area	0
BA1	Separation threshold between the L1# counting area and the WBC	35
BA2	Separation threshold between the WBC and basophils	110
BA3	End of the basophil counting area	240

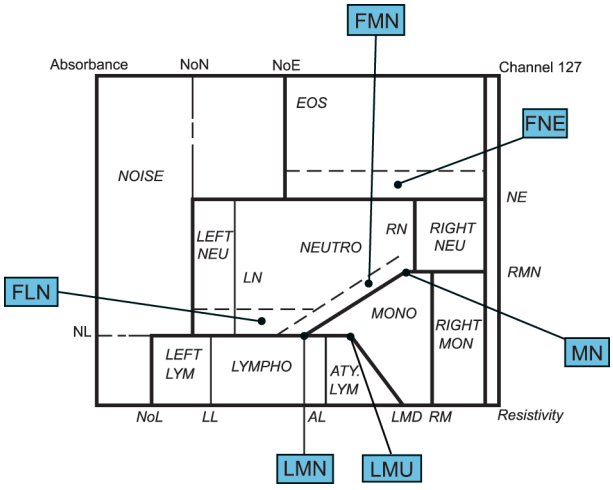
3.4.2. PLT

• The PL1 threshold is the number of the mobile channel that allows the calculation of the platelet population. It is automatically positionned.



3.4.3. LMNE matrix

• Each axis of the matrix (X and Y) is divided into 128 channels numbered from 0 to 127.
• 13 vertical indices (Y) and 13 horizontal indices (X) allow the user to locate these channels ten by ten. The first index of the matrix origin (at the bottom left) is the 0 channel, the fourth index of the matrix will be the channel 30 and so on.
• The threshold adjustment is expressed in channels.
• The threshold levels may be re-adjusted:
1 - To improve the separation between different cell populations which may vary according to the anti-coagulant in use or instrument internal adjustment.
2 - To modify the flag areas in one way or another to improve their detection sensitivity. In this case, the numeric adjustment of the concerned flag must also be readjusted too (see ***3.3. Flag Sensitivity***).
3 - To modify one or several matrix areas in order to define more precisely a specific population for research purposes.



DC thresholds (resistive) vertically shown on the matrix are adjustable by the user:

Threshold	Purpose	Standard	Low limit	High limit
NoL	Separation between Noise and Left Lymphocytes	22	0	LL
NoN	Separation between noise and Left Neutro	25	NoL	NoE
LL	Separation between Left Lymphocytes and Lymphocytes	30	NoL	AL
LN	Separation between Neutro and Left Neutro	35	NoN	LMN
NoE	Separation between the noise and Eosino	48	NoN Channel	127
LMN	Intersection dot between Lympho, Mono & Neutro thresholds	70	LN	LMU
AL	Separation between the Lympho and the Atypical Lympho	68	LL	LMU
LMU	Upper dot of the separation slope between Atypical Lympho & Mono	78	AL	LMD
LMD	Lower dot of the separation slope between Atypical Lympho & Mono	90	LMU	RM
MN	Upper dot of the separation slope between Mono & Neutro	90	LMN	RM
RM	Separation between Monocytes and Right Monocytes	118	LMD Channel	127
RN	Separation between Neutro and Right Neutrophils	118	MN Channel	127

AC thresholds (Absorbance) horizontally shown on the matrix are adjustable by the user:

Threshold	Purpose	Standard	Low limit	High limit
NL	Separation between Lymphocytes & Neutrophils	29	0	RMN
RMN	Separation between Right Mono & Right Neutrophils	51	NL	NE
NE	Separation between Neutrophils & Eosinophils	82	RMN Channel	127

The **FLN**, **FNE**, **FMN** thresholds indicate the width (in channel number) of the **LN**, **NE**, **MN** alarm areas:

Threshold	Purose	Standard
FLN	Channel number for the NL alarm area	2
FNE	Channel number for the NE alarm area	2
FMN	Channel number for the MN alarm area	2

- Move the cursor to the value to modify and enter the new value.

Press

to validate.

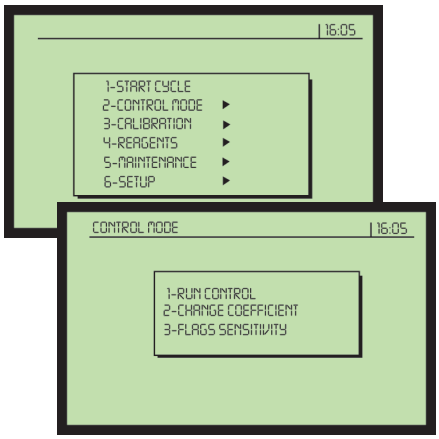
4. CONTROL MODE

• Control mode allows to run only *ABX* ***DIFFTROL*** control blood.

• Before running specimens it is recommended that the operator runs a «Normal» control blood to verify that the system is within acceptable limits.

• Prepare a «Normal» control blood according to the specific instructions detailed in the control blood package insert (temperature, mixing, etc...).

• Make sure control blood thresholds and alarms parametering is correct and corresponds to lot target values you are going to use: options «CONTROL MODE\CHANGE COEFFICIENT» and «CONTROL MODE\FLAGS SENSITIVITY» (report to the following paragraph for further details about parametering a new lot).



IMPORTANT

Risk of erroneous results if the control blood is not continuously mixed between each analysis.

• From menu «CONTROL MODE» select option «RUN CONTROL».

• QC samples identification can be entered into «QC» window «Next ID» field:

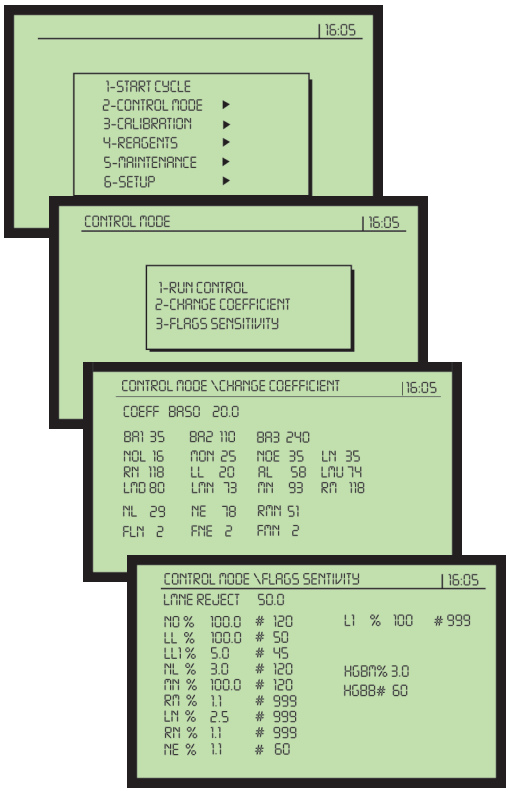
• Open the vial and start sampling.

NOTE

During control mode:
Results are printed with the title CONTROL.
Pathological limits, MIC & MAC alarms are disabled.
BASO curve is not printed.

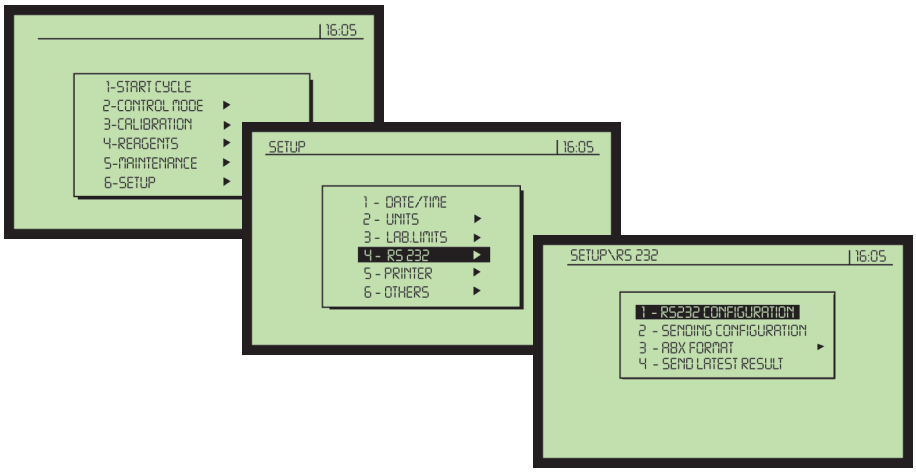
• In order to use *ABX* ***DIFFTROL*** control blood, it is necessary to adjust matrix thresholds and coefficients, according to the values delivered with the control blood lot you are going to use.

• Parametering is done through menus «CONTROL MODE\CHANGE COEFFICIENT» and «CONTROL MODE\FLAGS SENSITIVITY». Using ↓ key to select and modify value, press ↵ Enter key to validate.



5. DATA OUTPUT

• Several formats are available:
ARGOS: The **ARGOS** format allows the numerical result transmission only and the batch size is set to 406 characters for one result.
ABX: The **ABX** format allows the size of the transmitted data batches to be varied. Histograms, thresholds and matrix are available from the connection.
WORKS: The **WORKS** format enables the user to received data on a spreadsheet program from the RS232 port.



• This function allows the user to set up the RS232.

Connection mode

Two modes are available on *ABX PENTRA 60*:
• An unidirectional mode that performs the data transmission towards the main laboratory computer.
• A bidirectional mode which is performed acccording three different steps:
- Communication initialization
- Results transmission
- End of communication.

Protocol

XON/XOFF protocol controls data flow between *ABX PENTRA 60* and main laboratory computer.

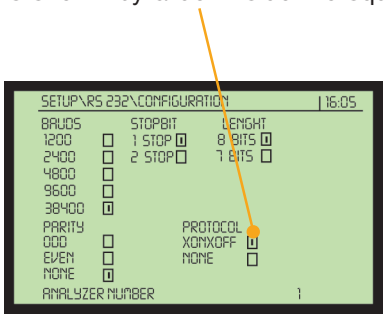
WARNING

Any modification of the instrument's DATA OUTPUT setup has to be done in agreement with the technician in charge of the main computer system. (the password is required to access to the RS232 menu)

5.1. RS232 Configuration

• Choose the baud rate, the parity, the character length, the stop bit and the protocol as follows:

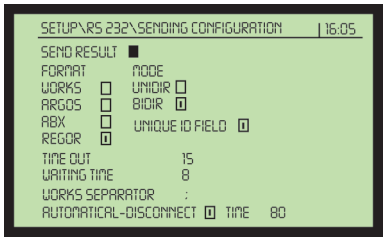
• Move the cursor to the parameter to be selected and press . : the selection is shown by a dot inside the square.



• Press Enter to validate.

5.2. Sending configuration

• This function allows the user to configure the format and the mode.



NOTE

The ABX format can be selected if the data transmission (length) is done on 8 bits.

5.3. ABX format

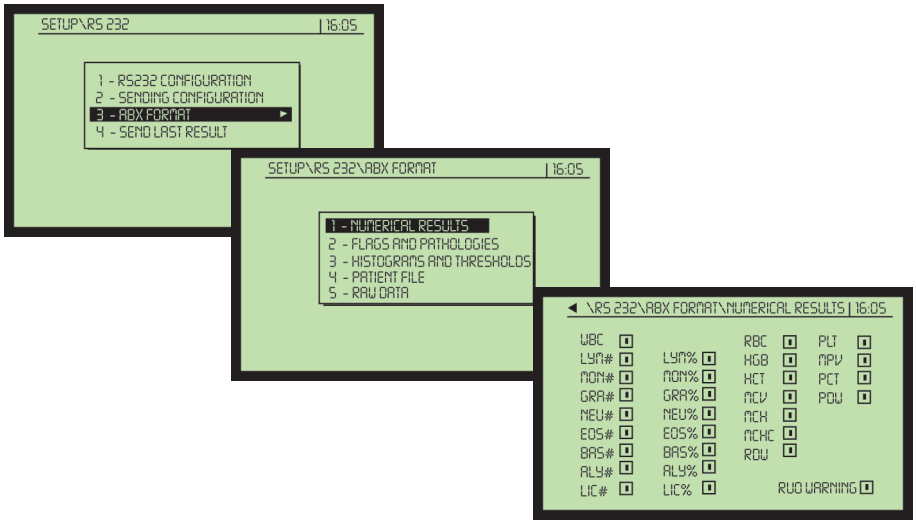
• A selection of transmitted parameters among numeric values, alarms and pathologies, curves and thresholds and patient files can be carried out (this selection only concerns **ABX format**).

5.3.1. Numerical values

• To select the hematologic parameters to be transmitted press 1. The «*Numerical values*» menu is opened.

• The dot inside the square displayed in front of the parameters indicates that these ones will be sent out.

• If some of them are not needed, delete the corresponding dot by means of .



NOTE

The «RUO» parameters are PCT, PDW, LIC and ALY. In «US mode» (setup by an approved HORIBA ABX technician at the instrument installation), the RUO parameters are automatically erased. In this mode, if the user has enabled these parameters from the menu «SETUP/OTHERS/IDENTIFICATION MODE», a warning message will be systematically printed out and sent to the host.

5.3.2. Flags and pathologies

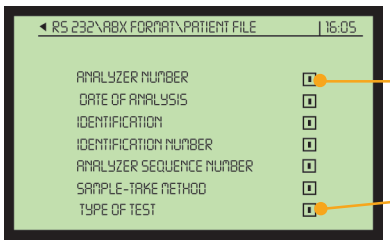
• As for «***5.3.1. Numerical values***», select the alarms and the pathology messages to be sent out by means of the window «*Flags and pathologies*» pressing **2**.

5.3.3. Histograms and thresholds

• As for «***5.3.1. Numerical values***», select the histograms and the thresholds to be sent out by means of the window «*Histograms and Thresholds*» pressing **3**.

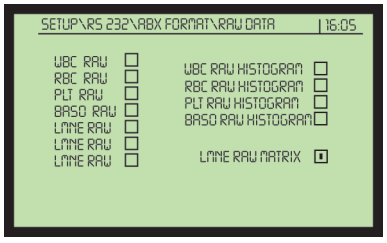
5.3.4. Patient file

• As for «***5.3.1. Numerical values***», select the patient file data to be sent out by means of the window «*Patient File*» pressing **4**.



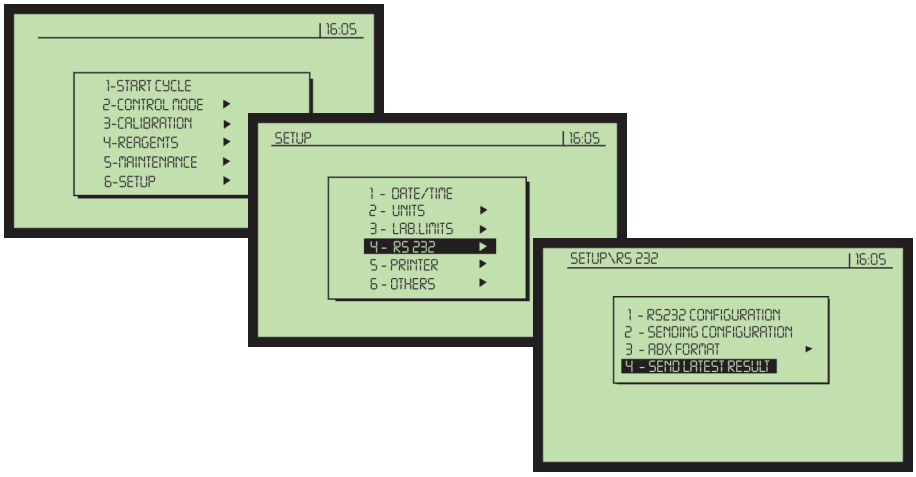
5.3.5. Raw values

• As for «***5.3.1. Numerical values***», select the Raw values to be sent out by means of the window «*Raw values*» pressing **5**.



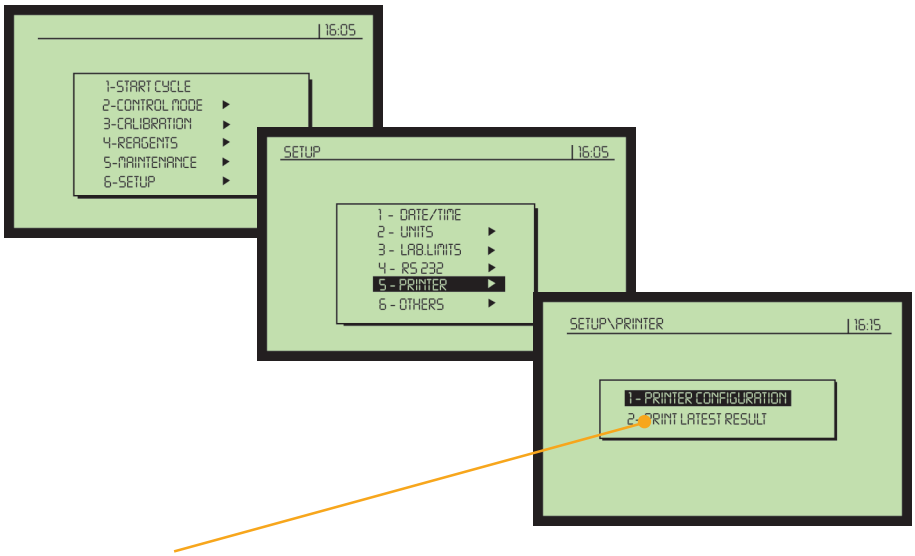
5.4. Send latest result

• Once the RS232 has been configured as described above, press **4** to send out the latest result to the main laboratory computer via the RS232 ouput.



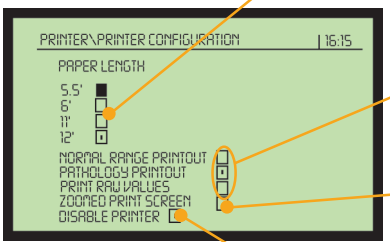
6. PRINTER

6.1. Print latest result



- Press **2** to print out the latest result configured as described below.

6.2. Printer configuration



Selection of the paper length:

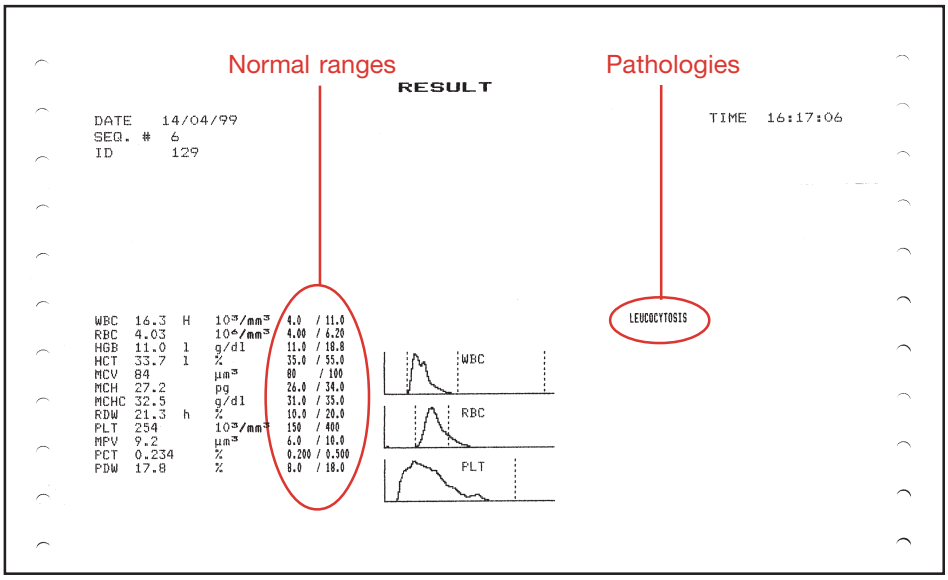
- Move the cursor by means of ↓ and ↑ to the correct length.
- The selection is performed when a dot appears in the square pressing . Factory adjusted to 12 inches.

Normal range, pathology printout, raw values:
(see result printout below)
Pathologies and normal ranges are printed out when a dot appears in both squares. Raw values of the counts can also be printed out.

Zoomed printed screen:

If enabled the screen printed out by means of the Print key will be zoomed.

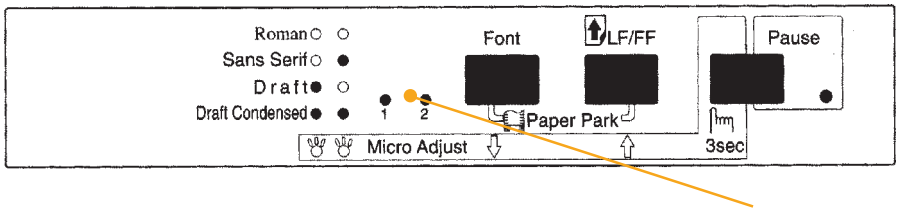
Disable printer:
When selected, the results are not printed out and no print alarm is triggered.



CAUTION

Character selection for the use on the ABX PENTRA 60 (see printer user manual for details).

- The printer must be configured in* Draft *mode.



- Pressing the FONT key will change the states of both LEDs 1 and 2 (swiched off or lit).

- To configure the* Draft *mode press the FONT key until the LED 2 is lit and the LED 1 is switched off.

7. OTHERS

7.1. Calibration

7.1.1. CV% limits

• This menu gives the variation coefficients **upper limits** for each parameter used in the calculations of the variation coefficients.

• When calibration CV results are not within these ranges, a flag «**H**» is triggered next to the CV value (see *Chapter 3. Specimen Run & Results* ***Calibration***).

• Move the cursor to the value to modify and enter the new value. Press Enter to validate.

• The factory values are shown on the below chart table:

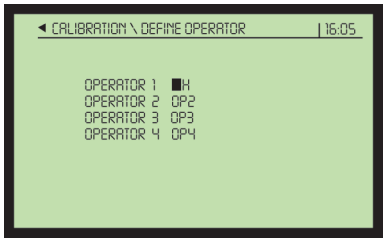
CV% Limits	Standard	min	max
WBC	2	1	3
RBC	2	1	3
HGB	1	1	2
HCT	2	1	3
PLT	5	3	7

7.1.2. Define Operator

• An operator identification is associated to calibration operations (see *Chapter 3. Specimen Run & Results* ***Calibration***). This one is modifiable from the menu «*Operator*».

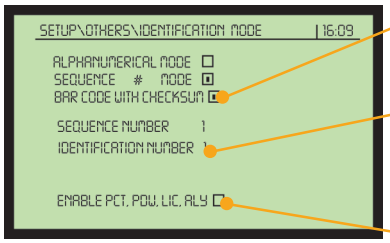
• 4 of them can be entered moving the cursor to the «*OP1*», «*OP2*», «*OP3*», «*OP4*» fields. Press Enter to validate.

• The selection of the operator is performed directly in the Calibration menu.



7.2. Identification mode

• To select either the **alphanumerical mode** or the **Sequence # mode** (see *Chapter 3. Specimen Run & Results*), move to the required mode and press . The selection is performed when a dot appears in the square.



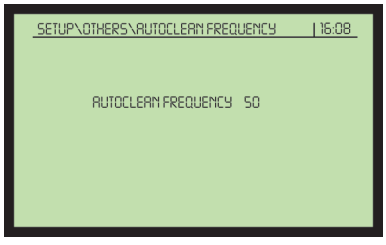
Barcode with checksum
Enables/disables the last character of the barcode (checksum).

Sequence # and identification # display
The sequence number indicates the number of analysis cycles from the beginning of the working day. This one is updated to 1 each new day.

Enable PDW, PCT, LIC, ALY
Enables/disables the RUO parameters (see ***5. Data output\5.3. ABX format***) for printing and sending towards the host.

7.3. Autoclean frequency

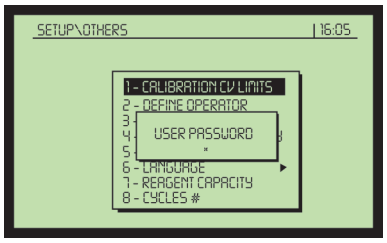
• Adjust the Autoclean frequency according to the number of analyses carried out (factory adjusted to 75 analysis cycles):
Minimum value: 1
Maximum value: 75



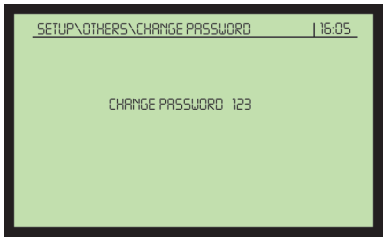
7.4. Change password

• The password is used to access the following functions:
- CALIBRATION / CHANGE COEFFICIENTS
- SETUP / LAB LIMITS / FLAG SENSITIVITY
- SETUP / LAB LIMITS / THRESHOLDS
- SETUP / RS 232
- SETUP / OTHERS / CHANGE PASSWORD
- SETUP / OTHERS / CALIBRATION CV% LIMITS
- FORCED CALIBRATION

• The original password is «123». To change it enter the previous password on this screen and press Enter.

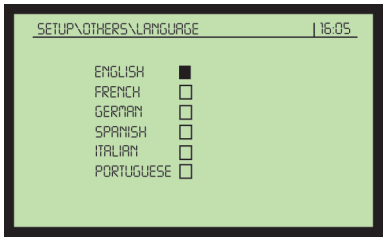


• Type in the new password on this screen and press Enter.



7.5. Change language

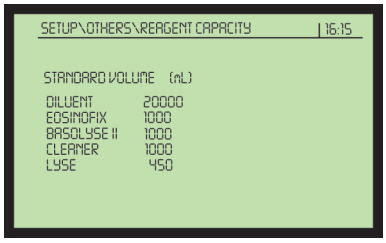
• The 6 following languages are available on the instrument:



• To modify the language, move the cursor to the wished language and press . to display a dot in the square. Press Enter to validate.

7.6. Reagent capacities

• This function indicates the initial volumes of each reagent bottle or container in order to calculate the consumption and the left reagent in each bottle.

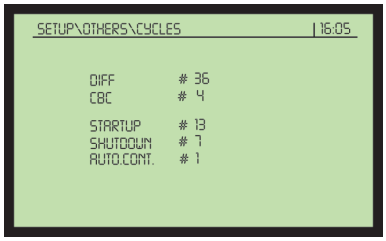


• Ranges are given in the below chart table:

Reagent	Standard capacity	Min	Max
DILUENT	20 000	1 000	30 000
CLEANER	1 000	100	5 000
EOSINOFIX	1 000	100	5 000
BASOLYSE 2	1 000	100	5 000
LYSE	400	100	2 000

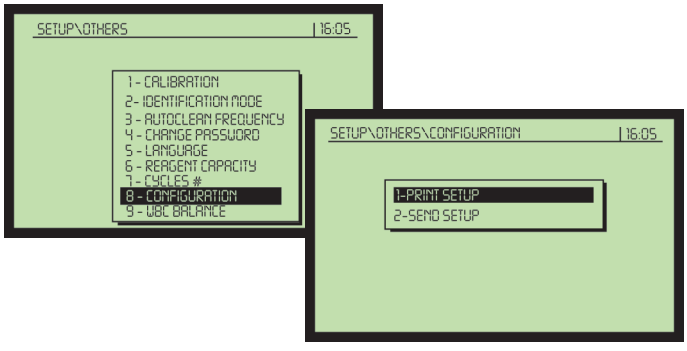
7.7. Cycle counters

• Shows the total number of analysis cycles (CBC & DIF mode) run on the instrument, as well as Startup, Shutdown and Autocontrol cycles.



7.8. Configuration

• Prints or sends (to the host) the setup of the instrument (coefficients, thresholds and flags values, internal parameters etc...).

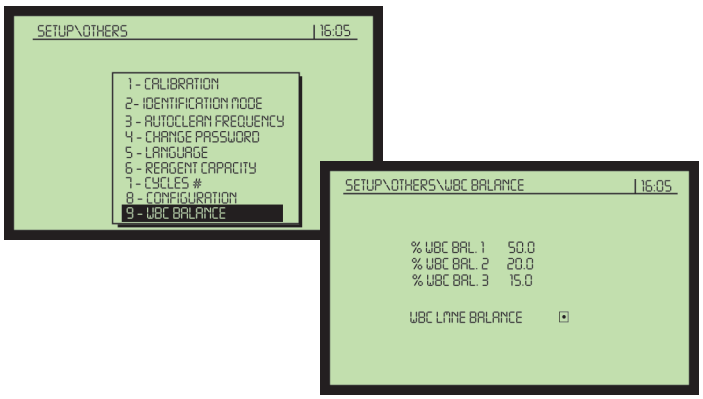


7.9. WBC Balance

• The meaning of these flags is described *Chapter 3. Specimen Run & Results* ***WBC Balance***.

• This window allows the user to enable or disable the WBC balance control (a dot in the square will enable the WBC balance).

• The triggering off thresholds of the balance flags are default values. They can not be modified by the user.



MAINTENANCE & TROUBLESHOOTING

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1. MAINTENANCE

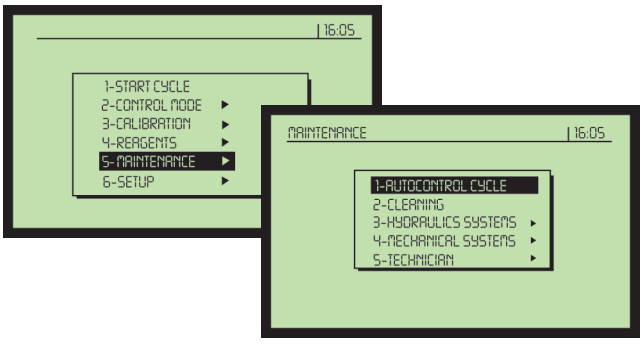
• One of the principle factors contributing to accurate and reliable results is a well-maintained instrument.
Several maintenance functions are available for the user to clean and check the instrument. On the chart table below, the maintenance cycle frequencies are indicated.

CYCLES	<100 ANALYSES PER DAY	>100 ANALYSES PER DAY
Shut down	1 per day	1 per day
Autoconcentrated cleaning	1 per month	2 per month

1.1. Autocontrol cycle

• This cycle is required after an emergency stop of the instrument or when a faulty operation has been detected.

A series of mechanical, hydraulic and electronic networks control is performed:
- General rinse.
- Control of the correct drains of the chambers.
- Initialization of the mechanical assemblies.

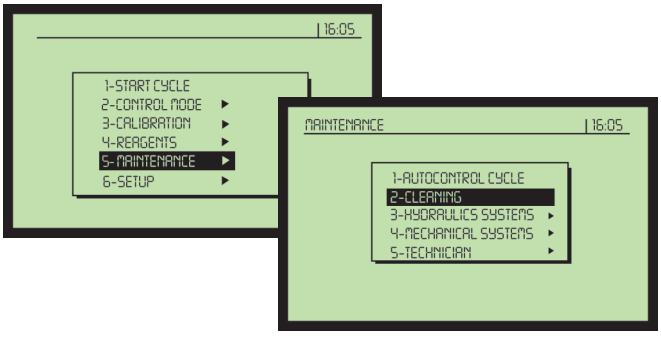


1.2. Cleaning cycle

• This cycle performs a chamber rinsing (as the «***Rinse / Chambers***» cycle) and it also primes reagent that could remained into the heating coil.

• After one hour off, the system automatically asks the user to run this cycle.

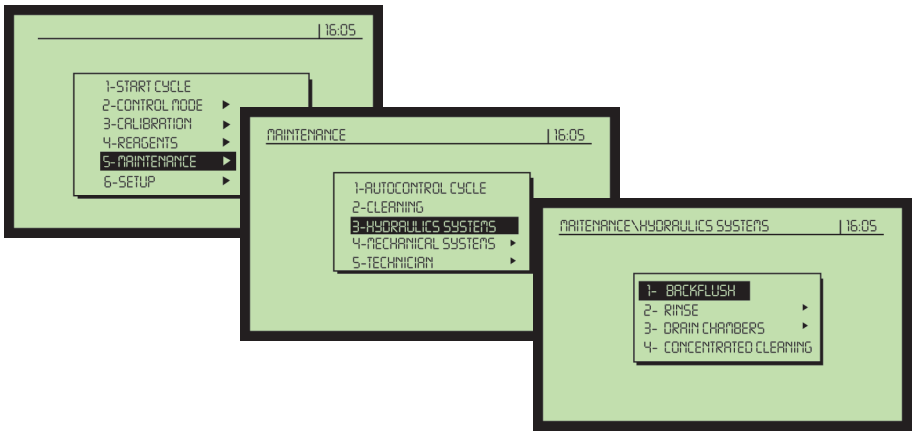
• User can run this cycle (after a stop shorter than an hour) by means of the beside menu.



1.3. Hydraulics systems

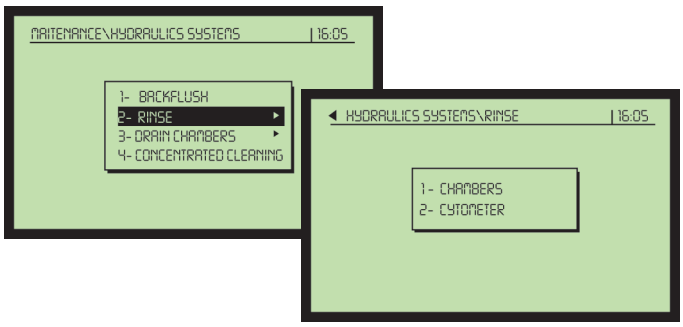
1.3.1. Backflush

• Delivers pressure through the apertures of the counting chambers to clean them in case of blockages.



1.3.2. Rinse

• *ABX* ***Diluent*** is sent to either chambers (pressing 1) or cytometer (pressing 2) to rinse out these parts separately.



NOTE

The cytometer rinse includes a sequence that removes bubbles that could remain inside the flowcell.

1.3.3. Drain chambers

Runs chamber draining cycle for the following parts:
- *Rinse*

Sampling probe rinsing chamber *(1)* drain
- *First dilution*

First dilution chamber *(2)* drain
- *LMNE*

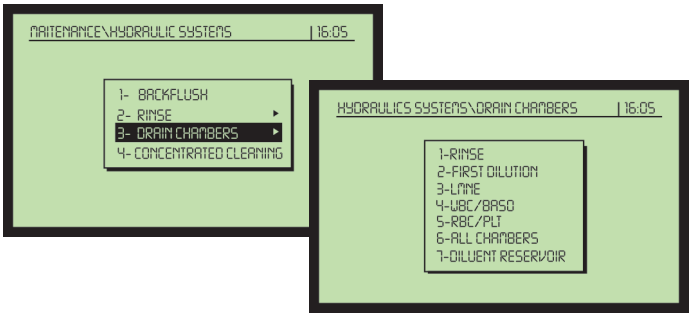
LMNE chamber *(3)* drain
- *RBC/PLT*

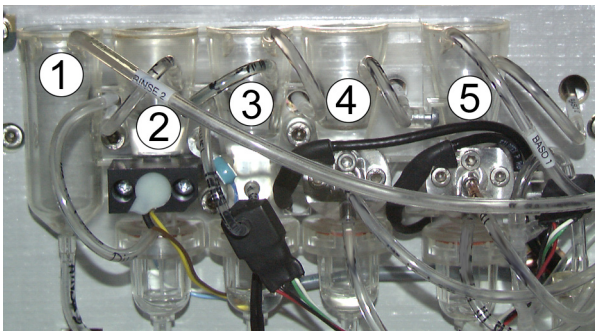
RBC/PLT chamber *(4)* drain
- *WBC/BASO*

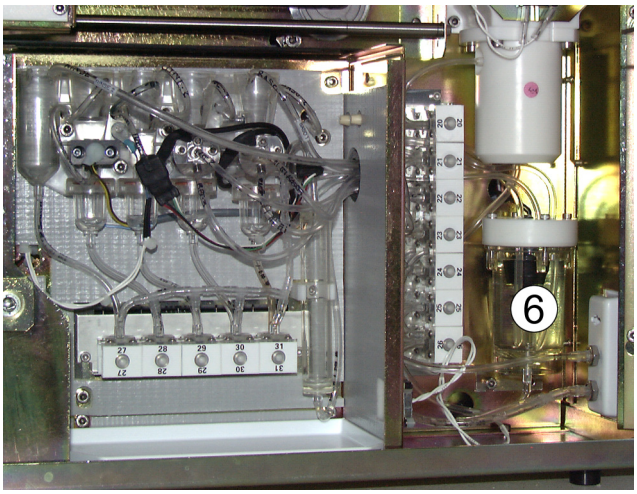
WBC/BASO chamber *(5)* drain
- *All chambers*

General drain
- *Diluent reservoir*

Diluent reservoir *(6)* drain

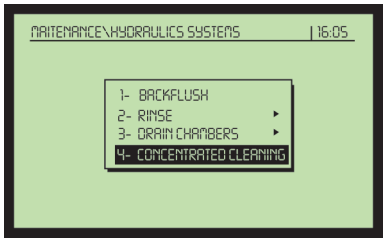






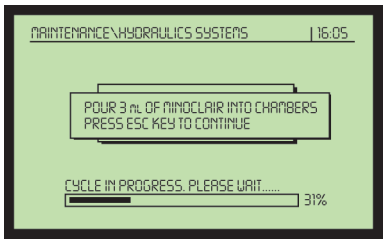
1.3.4. Concentrated cleaning

- Cleans the chambers with bleach.

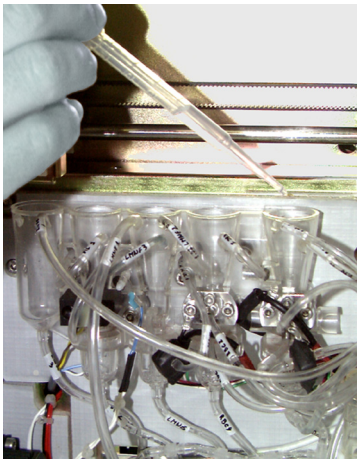


Procedure:

- Press Enter: a rinse cycle is run.
- Wait for the following message to be displayed.


MAINTENANCE\HYDRAULICS SYSTEMS | 16:05
POUR 3 mL OF MINOCLAIR INTO CHAMBERS
PRESS ESC KEY TO CONTINUE
CYCLE IN PROGRESS. PLEASE WAIT.....
31%

- Open the instrument pneumatic door.



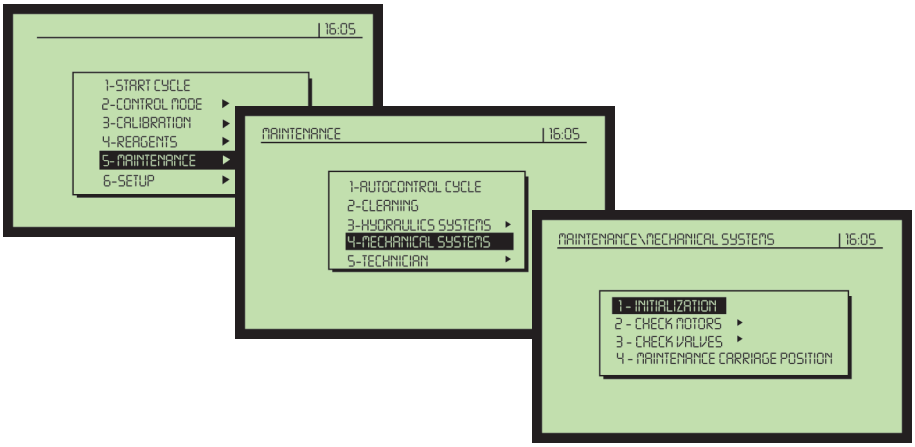
- Using 5ml syringe, pour 3ml of MINOCLAIR (or bleach diluted to 4° chloride) into each chamber and press ESC.

- Close the instrument door and wait for the instrument to complete the cleaning procedure (Concentrated cleaning duration around 5 mn).

1.4. Mechanical systems

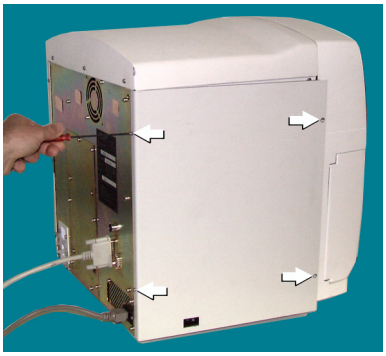
1.4.1. Initialization

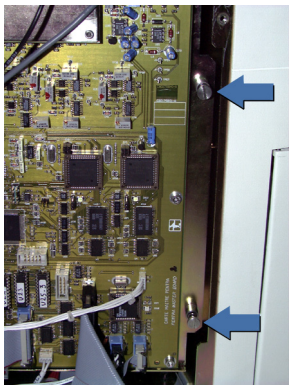
• All the mechanical assemblies (sampling probe, carriages, syringes...) return to their initial positions, i.e. the operating analysis positions.



1.4.2. Check motors

To control the correct operation of each motor:
• Switch off the instrument.
• Open the door (see on previous page) and the left cover of the instrument:

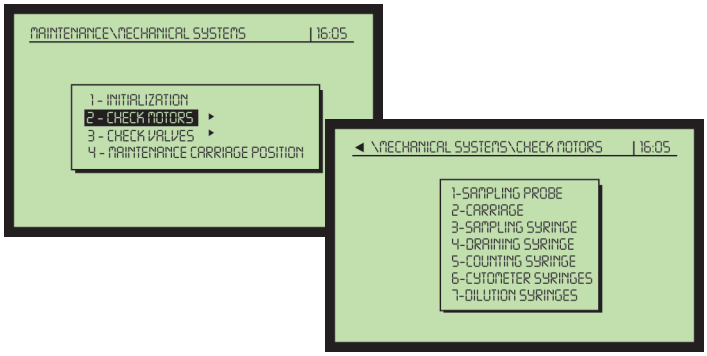




CAUTION

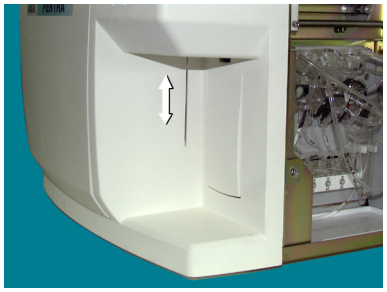
Be careful to the flat cables while opening the door!

• Loosen the 2 screws of the board support panel and open it.
• Once both sides of the instrument are open, switch on the instrument. Enter the check motors menu and control each motor pressing the corresponding number.

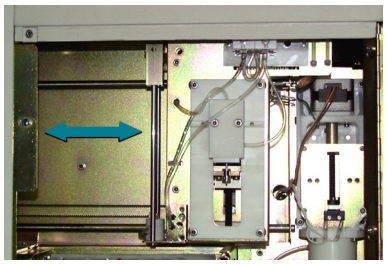


Right side of the instrument

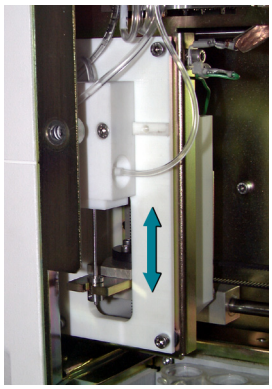
1. Sampling needle: Check the needle up and down operations. The movements should be smooth and regular.

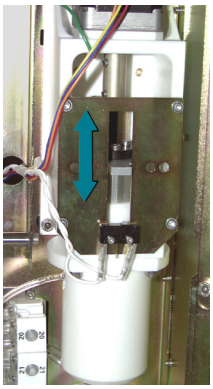


2. Carriage: Check the right and left movements of the carriage.



3. Sampling syringe: Check that the syringe up and down movements are smooth and regular.

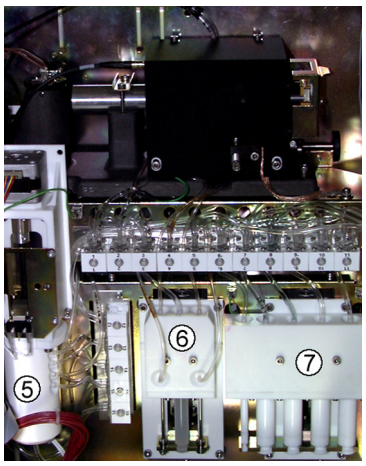




4. Draining syringe: Check the correct up and down movements of the syringe.

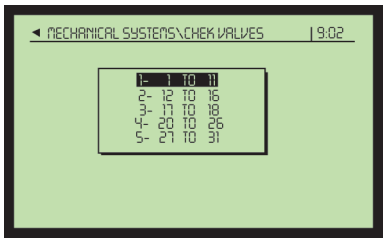
Left side of the instrument

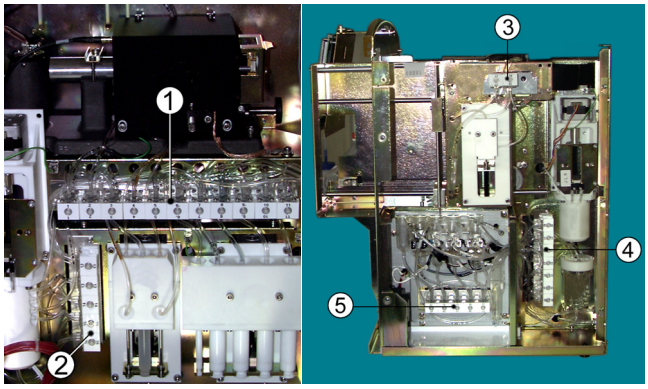
5. Counting syringe: Same operation check.
6. Cytometer syringes: Same operation check.
7. Dilution syringes: Same operation check.



1.4.3. Check valves

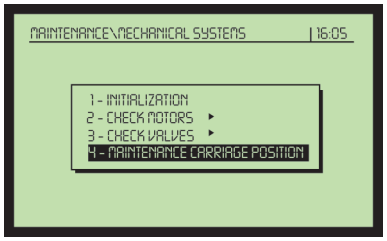
To control the correct operation of the valves:
- Open the door (see on previous pages) and the left cover of the instrument.
- The valves can be operated pressing the corresponding number of the assembly.
- Closely observe the valve operations; the movements have to be straight and regular.





1.4.4. Maintenance carriage position

• The probe goes up, the carriage comes under chambers and all stepper motors are freed for the user to do any maintenance.



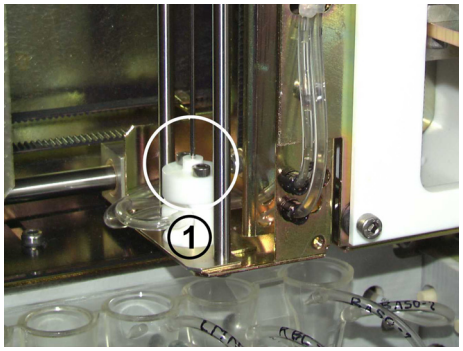
• To bring the probe back, press Enter key, once more.

1.5. Technician

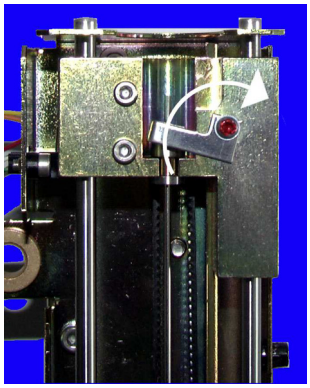
• Reserved to *HORIBA ABX* Field Service Engineer.

1.6. Sampling probe replacement

- Switch off the instrument and remove the power cable.
- Open the pneumatic access door (Right side of the instrument).
- Unscrew both fixation screws of the probe rinsing block on the carriage:

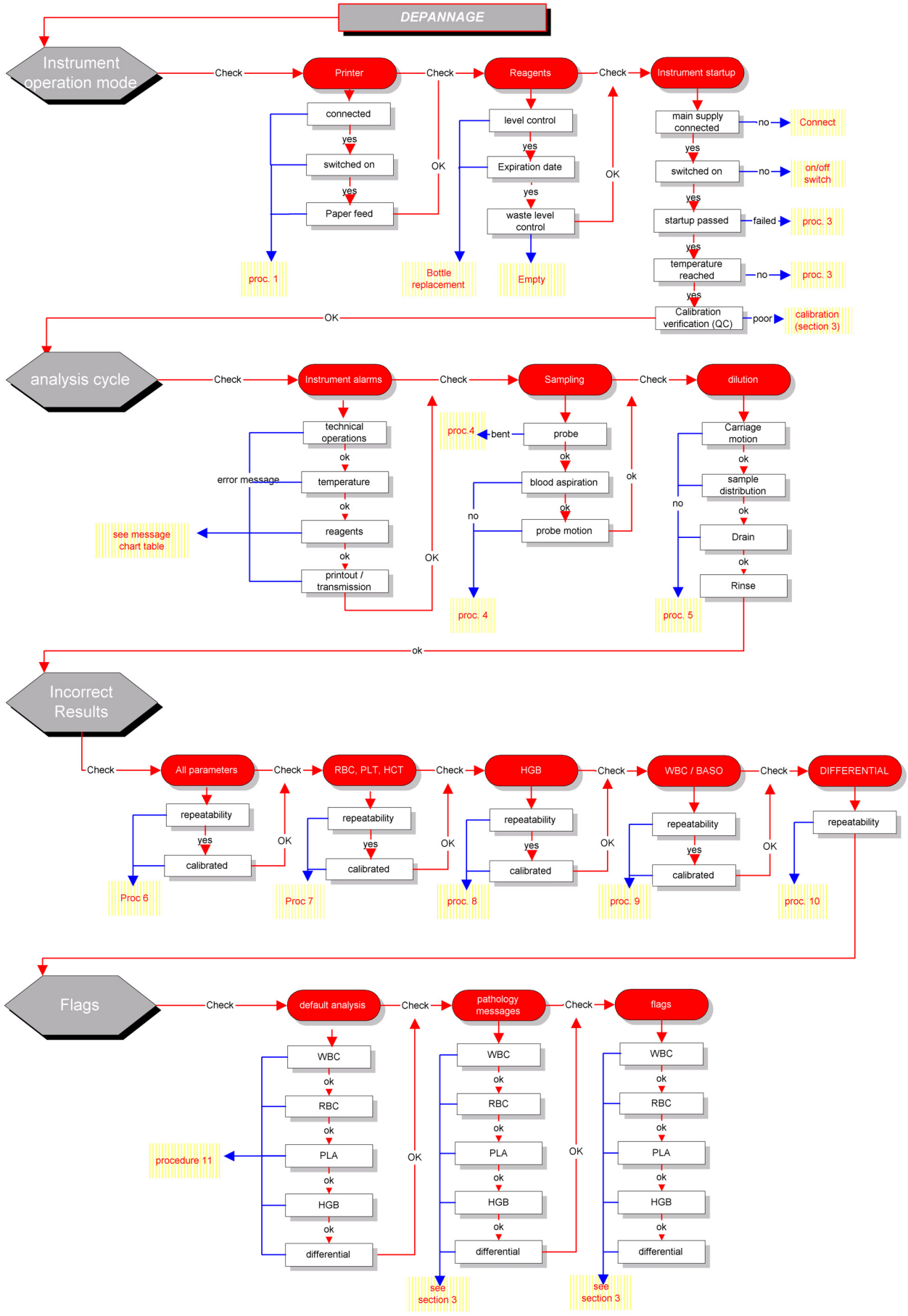


- Take out the probe and the rinsing block at the same time (lift the probe locker to free the probe). Replace the probe.



- Reassemble following previous steps backward.
- When startup is done check for that there is no leak.

2.TROUBLESHOOTING



2.1. Instrument operation mode

Procedure 1: Printer

- See *Printer user manual* to connect, to switch on/off or to feed paper.

Procedure 2: Reagents

- Bottle replacement (See *Chapter 3. Specimen Run & Results*).
- Waste container: empty and neutralize as recommended in section ***1. Specifications***.

Procedure 3: Instrument startup

Startup failed:

- Check the reagent expiration dates: replace bottle if necessary.
- Re-run a startup.
- Perform an auto-concentrated cleaning (see ***1.3.4. Concentrated cleaning***).

Temperature not reached:

- Wait for 5 minutes to reach the operating temperature.
- Call your *HORIBA ABX* representative Service Department.

Calibration verification out of the acceptable limits:

- Clean the system (see ***1.3.4. Concentrated cleaning***) and rerun the control.
- Run a new vial.
- Calibrate the instrument (See *Chapter 3. Specimen Run & Results*).

2.2. Analysis cycle

Error messages

Printer

Displayed message	Causes	User actions
The printer is disconnected, switched off or has not been selected	Printout operations disabled	- Switch on or - Press «ON LINE» or - See the printer's user's manual.
Defect on printer, make sure there is paper	Printout operations disabled	- Feed paper or - See the printer's user's manual.
Printer being used, action impossible	The printer operates yet	Wait for the current printout to complete and restart the request.

Transmission

Displayed message	Causes	User actions
No ENQ character received on RS232 No ACK character received on RS232 Internal error on RS232 Write error RS232 Timeout overflow on RS232 CRC error Instrument number error Message lenght error Receiving data error	Defect on transmission operations.	Check the RS232 configuration Menu «SETUP / RS 232/ RS232 CONFIGURATION». Call HORIBA ABX representative service department.

Calibration

Displayed message	Causes	User actions
Access denied	Incorrect pasword entered by the operator	Re-type the password.
Data not saved, value out of range	Incoherent value entered by the operator	Re-type in the item.
Illegal date	Incoherent date entered by the operator	Re-type in the date.
Minimum tagged CBC incorrect, at least 3	Selected results for calibration calculation < 3	Select at least 3 results.
Max num. done, start cycle refused	11 results are already recorded in the calibration table	Perform the procedure described section 3 to change coefficients or to exit the calibration menu.

Miscellaneous

Displayed message	Causes	User actions
Emergency stop, Run an autocontrol	Blocked motor Incorrect Drains Thermal door opened	Control the motor operations: Menu «MAINTENANCE / MECHANICAL SYSTEMS / CHECK MOTORS».
.....not reaching home	Blocked motor	
Thermal door opened	Open during a cycle	Close the door and rerun the cycle.
Illegal time	Incoherent time entered by the operator	Enter the correct time.
Data not saved, value out of range	Incorrect value entered by the operator	Enter a correct value.
User password	Password required to carry out an operation	Enter the password.
Enter an identification	To run an analysis in alphanumerical mode, the identification is mandatory	Enter the identification as described section 3 of this manual.

Reagents

Displayed message	Causes	User actions
No diluent, check level	Diluent reservoir empty	Replace the diluent container (menu «REAGENTS / LEVEL-CHANGE»).
Reagent low level (reagent name)	None	Replace the bottle (menu «REAGENTS / LEVEL-CHANGE»).
Reagent low level	Message triggered at the end of the Startup	Control the reagent levels or/and replace it.
Drain sensor time out	Chamber and/or syringe draining problems	Call HORIBA ABX representative Service Department.
Transfer sensor time out	Transfer problem with LMNE matrix sample	Call HORIBA ABX representative Service Department.

Temperature

Displayed message	Causes	User actions
Temperature out of range	Thermic regulation problem	Call HORIBA ABX representative department.

Procedure 4: Sampling

Sample probe:

• Check the motion of the probe (menu ***maintenance/mechanical systems/ check motors/sample probe***).
• Open the pneumatic access door.
• Run an analysis cycle on blood.
• Control the specimen aspiration (blood delivered in the chambers).
• Check the probe is not bent.

Procedure 5: Dilution

Carriage motion:

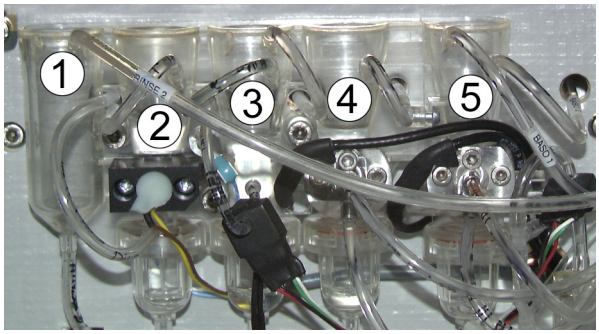
• Check that hydraulic operations appear to work properly (reagent level in each chamber, carriage motion).

Sample distribution:

• Run an analysis cycle and check that the specimen distribution is performed correctly into the chambers.
A probe rinse is previously carried out in the rinse chamber (1) (blood appears in this chamber).
The first specimen is delivered to the first dilution chamber (2) (brown colour), the second to the WBC/BASO chamber (5) (clearer)
and the third one to the LMNE chamber (3) (the darkest). Check that bubbling is provided to these chambers once the specimen have been diluted.

Drain and rinse:

• Check the chambers are drained and rinsed.
If operations are faulty, identify the source of the malfunction and call your *HORIBA ABX* representative Service Department.



2.3. Results

Procedure 6: All parameters

Repeatability (according to the CV Specifications see section 1):

- Is the instrument non repeatable on all parameters? If not perfom directly the procedure corresponding to the non repeatable parameter.
- If all parameters are not repeatable:
- Visually check that the sampling operation appears to be correct.
- Control the sampling syringe operations (see ***1.3. Mechanical systems***).
- Control the counting syringe operations (see ***1.3. Mechanical systems***).
- Perform an autoconcentrated cleaning.
- If all these operations appear to be correct, call your *HORIBA ABX* Representative Service Department.

Calibration:

- If the system appears to be operating properly, fresh uncontaminated reagents are being used and the precision is within the specifications, the *ABX PENTRA 60* may need a calibration as described *Chapter 3. Specimen Run & Results*.

Procedure 7: RBC, PLT, HCT

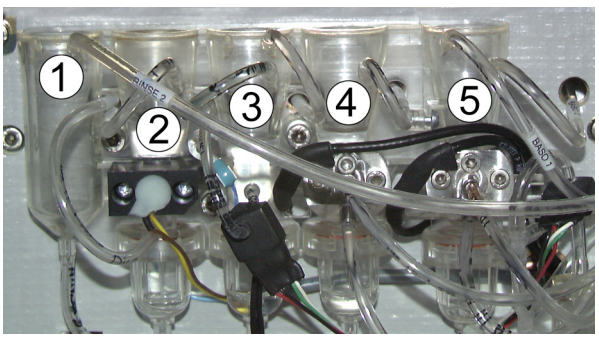
Repeatability:

If RBC, PLT & HCT are not repeatable:

- Check the second dilution is carried out correctly (sample from the chamber 2 to the chamber 4).
- Check the Bubbling in the RBC/PLT chamber (4) once the dilution is carried out (the dilution remains transparent).
- Perform an autoconcentrated cleaning.
- If all these operations appear to be correct, call your *HORIBA ABX* Representative Service Department.

Calibration:

- See procedure 6.



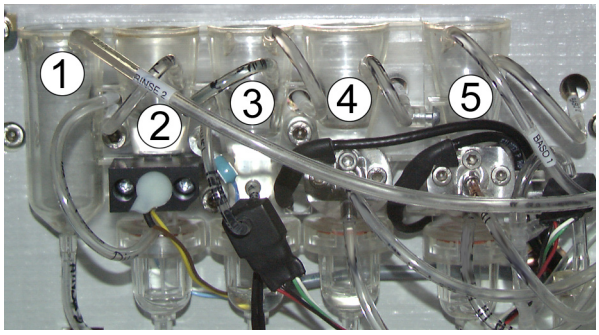
Procedure 8: HGB

Repeatability:

- Is the instrument non repeatable on HGB?
- Run a analysis cycle.
- Check the dilution colour in the chamber (2).
- «Milky» when the sample is first delivered to the chamber, then brown transparent when Lyse is injected.
- Perform an autoconcentrated cleaning.
- If this does not correct the HGB count, call your *HORIBA ABX* Representative Service Department.

Calibration:

- See procedure 6.



Procedure 9: WBC/BASO

Repeatability:

- Is the instrument non repeatable on WBC/BASO?
- Perform an autoconcentrated cleaning.
- If this does not correct the WBC/BASO count, call your *HORIBA ABX* Representative Service Department.

Calibration:

- See procedure 6.

Procedure 10: Differential

Repeatability:

- Is the instrument non repeatable on differential?
- Perform an autoconcentrated cleaning.
- If this does not correct the WBC/BASO count, call your *HORIBA ABX* Representative Service Department.

2.4. Flags

Procedure 11: Default analysis

WBC:

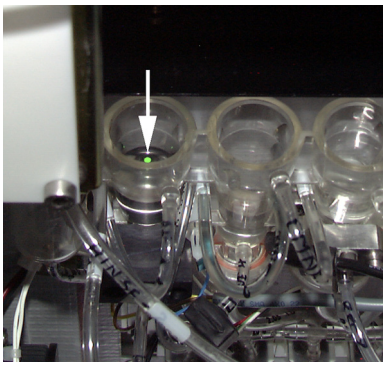
- Perform an autoconcentrated cleaning.
- Re-run the specimen.
- Check the operation of the liquid valve <23> and <14> (opening and closing during the cycle). If defective, replace the valve.
- If it does not correct the WBC results, call your *HORIBA ABX* representative Service Department.

RBC, PLT:

- Perform an autoconcentrated cleaning.
- Re-run the specimen.
- Observe the operation of the liquid valve <14> (opening and closing during the cycle). If not replace the valve.
- If it does not correct the RBC or PLT results, call your *HORIBA ABX* representative Service Department.

HGB:

- Is the HGB LED illuminated when the system power is on? If not call your *HORIBA ABX* representative Service Department. If it is continue.
- Perform an autoconcentrated cleaning.
- Re-run the specimen.
- If it does not correct the HGB results, call your *HORIBA ABX* representative Service Department.

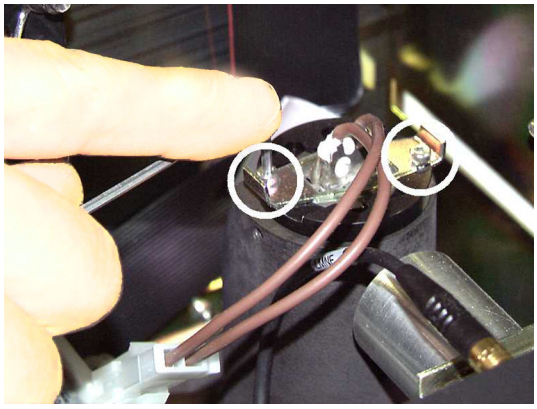


Differential:

- Control that the lamp is still illuminated when the instrument is on. If not replace it as described below.
- Run a cytometer rinse (menu ***maintenance/hydraulics systems/rinse/ cytometer***).
- Re-run the specimen.
- If it does not correct the LMNE results, call your *HORIBA ABX* representative Service Department.

Lamp replacement:

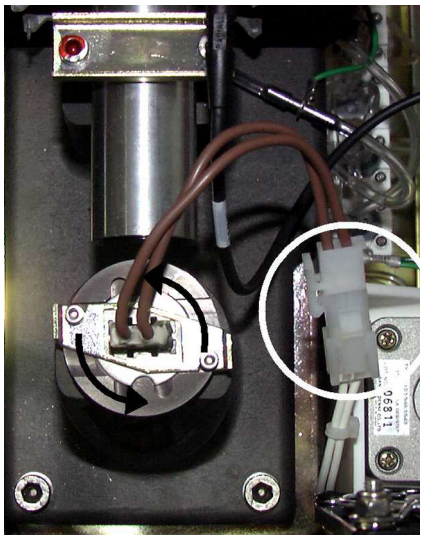
- Switch off the instrument.
- Open the cover.
- Disconnect the lamp supply.
- Unscrew lamp fixation screws (few turns).



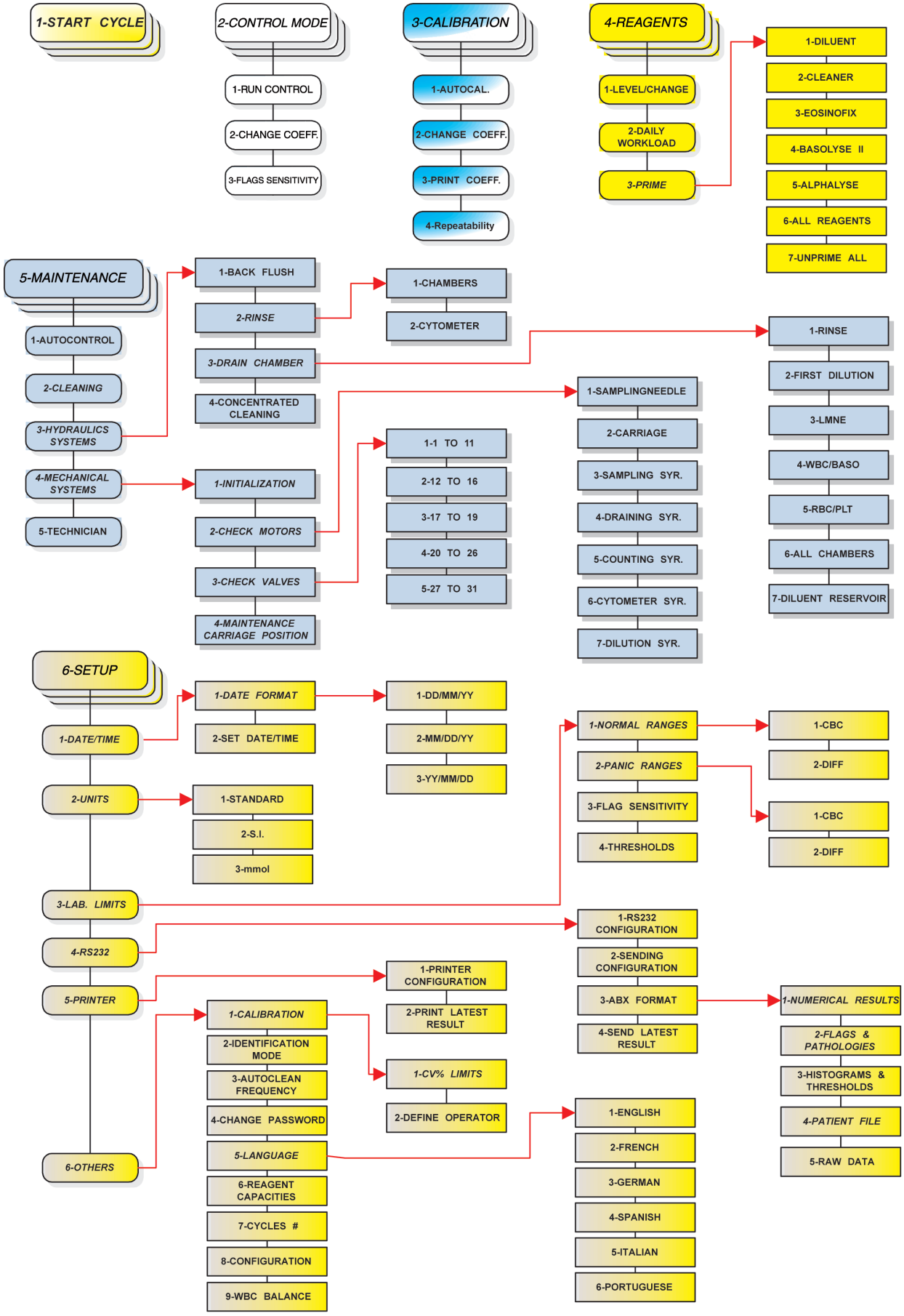
WARNING

Wait for the lamp to cool down before handling it!

- Turn the lamp and remove it.
- Replace the lamp by a new one.
- Put back the fixation system and block the screws.
- Reconnect the lamp supply.



3. MENU OVERVIEW



ANNEX

GLOSSARY 1-2
LIST OF ABBREVIATIONS 1-4
PNEUMATIC DIAGRAM 1-6

GLOSSARY

	Definition
accuracy	Ability of the instrument to agree with a predetermined reference value at any point within the operating range; closeness of a result to the true (accepted) value
agglutination	Clump
background count	Measure of the amount of electrical or particle interference
blank cycle	Runs diluent through the system to clean it out
calibration	A procedure to standardize the instrument by determining its deviation from calibration references and applying any necessary correction factors
calibration factors	These are correction factors that the system uses to fine-tune instrument accuracy
calibrator	A substance traceable to a reference method for preparation or material used to calibrate, graduate, or adjust measurement
carryover	The amount, in percent, of blood cells remaining in diluent following the cycling of a blood sample
cell control	A preparation made of human blood with stabilized cells and surrogate material used for daily instrument quality control
characteristics	See performance characteristics
coefficient of variation	An expression in percent of data (SD) spread related to the mean CV%=(SD/mean)x100
control	A substance used for monitoring the performance of an analitycal process or instrument
CV	See Coefficient of variation
default	An original factory setting
expiration date	The last day that you can use that specific lot number of reagent, control or calibrator
fL	Abbreviation for femtoliter
femtoliter	One quadrillionth (10^{15}) of a liter
field	An area on a screen for entering data
flags	On printouts or screens, letters or symbols that appear next to parameter results to indicate specific conditions
linearity	The ability of an instrument to recover expected results (reference values or calculated values) for such parameters as WBC, RBC, Hgb and Plt, at varying levels of concentration of these parameters within specified limits
lot number	A manufacturer's code that identifies products such as reagents, controls or calibrators

mean	Arithmetic average of a group of data
operating range	Range of results over which the instrument displays, prints and transmits data
parameter	A component of blood that the instrument measures and reports
performance characteristics	Actual performance of the instrument
performance	Targeted performance of the instrument based on established ranges and parameters specifications
quality control (QC)	A comprehensive set of procedures a laboratory establishes to ensure that the instrument is working accurately and precisely
reproducibilty	This procedure checks that the system gives similar results (within established limits) every time it measures the same sample
SD	A measure of variation within a group samples or within a population (standard deviation)
shutdown cycle	Cleans the instrument's fluidic lines and apertures to help prevent residue buildup
specifications	See performance specifications
startup cycle	Ensures that the instrument is ready to run; includes performing a background test
verification	Procedure to analyze cell controls or whole blood with known values to determine if your results are within the acceptable range
whole blood	Non-diluted blood; blood and anticoagulant only

LIST OF ABBREVIATIONS

Abbrevitaion	Meaning
µL	microliter
µm	micrometer
ACD	acid-citrate-dextrose
ALY	Atypical Lymphocyte
BAS or BASO	basophil
bps	bit per second
CBC	cell blood count
Cl	chlorine
cm	centimeter
CV	coefficient of variation
DHSS	double hydrodynamic sleeving
diff	differential
dL	deciliter
EDTA	ethylenediaminetetraacetic acid
EOS	eosinophil
fL	femtoliter
ft	foot or feet
g	gram
Gb	gigabyte
Hct	hematocrit
Hgb	hemoglobin
Hz	hertz
L	liter
lb	pound
LED	light-emitting diode
LIC	Large Immature Cell
LYM	lymphocyte
m	meter
mb	millibar
Mb	megabyte
MCH	mean corpuscular hemoglobin
MCHC	mean corpuscular hemoglobin concentration
MCV	mean corpuscular volume
MDSS	multi distribution sampling system
MHz	megahertz
mL	milliliter
mm	millimeter

MON	monocyte
MPV	mean platelet volume
MSDS	material safety data sheet
n	number
NEU	neutrophil
nm	nanometer
Pct	Plateletcrit
PDW	Platelet Distribution Width
Plt	platelet
RBC	red blood cell
RDW	red distribution width
SD	standard deviation
VA	voltampere
Vac	volt alternatif current
WBC	white blood cell

LV1: Differential diluent - Selects flow cell sheath 2 or LMNE chamber
LV2: Differential diluent - Selects flow cell sheath 1 or sheath 2
LV4: Flow cell sample supply - Opens pathway from the LMNE chamber to the flow cell
LV5: Flow cell sample injector - Opens waste path for sample injector syringe
LV10: Diluent output control - Routes diluent to probe rinse block or heating coil
LV12: Rinse output control - Selects rinse to probe rinse block or WBC/BASO chamber
LV14: RBC/PLT count valve - Opens vacuum count line for RBC chamber
LV20: Waste syringe vent - Opens waste vent (through the rinse chamber)
LV21: Sweep flow diluent - Routes diluent to heating coil or sweep flow
LV22: Diluent bath select - Routes diluent (via heating coil) to Hgb or RBC chamber
LV23: WBC/BASO count vacuum - Routes vacuum direct or through RBC/PLT counting head
LV24: Flow cell drain: Opens path from flow cell output to LMNE chamber for drain

TRANSLATION Inch To mm (int. diam.)			
0.010"	0.25 mm	0.045"	1.14 mm
0.020"	0.50 mm	0.051"	1.30 mm
0.025"	0.64 mm	0.060"	1.52 mm
0.030"	0.76 mm	0.081"	2.05 mm
0.035"	0.89 mm	0.090"	2.28 mm
0.040"	1.02 mm	0.0100"	2.54 mm

C: CRISTAL TUBE
: TYGON TUBE

+ DRAIN
+ BUBBLING
+ LMNE TRANSFERT
+ PROBE WIPE

+ COUNT
+ DILUENT FILL
(TANKER FILLING)

	Description	Chk	By	Date	ECO	Rev
D	Rectify the length of the tubing between LV23-1 and RBC bath-3: 1.52 220 was 200.	CTH	ADC	04/23/01	86	
C	2.54 170 and 2.54 140 was 2.05. Add 3 sleeves. 1.52 350 was 290. ECO 4868: 1.85 10 was 11. ECO 4582: 2.05 15 was 20 and 2.05 50 was 40.	CTH	ADC	02.22.2000	4921	
B	Tube 1.52 260 was 250 (Reagent heater-1 / Valve 22-2). ECO4583: Add waste and diluent external tubes.	CTH	ADC	07.12.1999	4633	
A	Add sleevings on external sheath circuit. Creation according P60 prototype diagram PNEU-D. Update ECO4310 and ECO4411-suite.	CTH	ADC	05.11.1999	4399	

Designation: **HYDROPNEUMATIC DIAGRAM PENTRA 60 OT**
Standard tolerance: - | Roughness: -
Material: -
Treatment: -
Coating: -
N° : **NML001A** | Sheet: **S** | Drw size: **A2** | Scale: - | Dim. in millimetres

DIAGNOSTICS

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